



Sedentary behaviour and cardiometabolic health in adolescents: role of diet

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**Sedentary Behaviour and
Cardiometabolic Health in Adolescents:
Role of Diet**

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BExSc&Nutr; BNut(Honours)

Submitted for fulfilment of the award of
Doctor of Philosophy

Deakin University

6th November 2017



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Abstract

Background: Television (TV) viewing is consistently linked with chronic conditions, such as obesity and metabolic syndrome, in adolescents. However, associations with other sedentary behaviours, such as total sedentary time and bouts of sedentary time, are less clear. Given dietary intake is associated with TV viewing, dietary intake could partially explain why TV viewing is consistently related to cardiometabolic health outcomes.

Aim: The overall aim of this thesis is to explore the role of dietary intake in the relationship between various sedentary behaviours and cardiometabolic risk markers in adolescents. To do this, four studies were conducted to specifically address four main objectives: *Study 1*) Examine the cross-sectional mediating role of dietary intake on the relationship between self-reported TV viewing, body mass index (BMI) and metabolic syndrome in U.S. adolescents; *Study 2*) Examine the cross-sectional mediating role of dietary intake on the relationship between total volume and bouts of sedentary time (accelerometry measured), BMI and metabolic syndrome in U.S. adolescents; *Study 3*) Examine the cross-sectional and prospective mediating effects of dietary intake on the relationship between self-reported TV viewing, total volume and bouts of sedentary time (accelerometry measured) and BMI in Australian adolescents; and *Study 4*) Examine the role of dietary intake (high-energy diet *versus* standard-energy diet) on the relationship between uninterrupted sitting, sitting with resistance-type activity breaks and postprandial glucose in apparently healthy Australian adolescents.

Methods: For *Studies 1, 2* and *3*, mediation analyses were conducted to examine the role of various dietary mediators (e.g. total energy intake, fruit and vegetable intakes,

consumption and frequency of discretionary foods and sugar-sweetened beverages, and diet quality) on the relationship between different types of sedentary behaviours (e.g. TV viewing, total sedentary time and bouts of sedentary time) with BMI and metabolic syndrome. For *study 4*, participants completed a four-treatment cross-over pilot trial: 1) uninterrupted sitting and consumption of high-energy diet; 2) sitting with breaks and consumption of high-energy diet; 3) uninterrupted sitting and consumption of standard-energy diet; and 4) sitting with breaks and consumption of standard-energy diet. Linear mixed models were used to examine differences between postprandial glucose area under the curve (measured via a continuous glucose monitoring system) for each of the four conditions.

Results: The findings from *Study 1* showed sugar-sweetened beverage consumption and fruit and vegetable intake partially mediated the relationship between TV viewing and metabolic syndrome in adolescents by 8.7% and 4.1%, respectively. However, no significant mediation effects were observed for the relationship between TV viewing and BMI. In both *Studies 2* and *3*, none of the dietary variables examined mediated the relationship between total volume and bouts of sedentary time with BMI and metabolic syndrome, both cross-sectionally (*Studies 2 and 3*) and prospectively (*Study 3*). Lastly, findings from *Study 4* showed, compared to the uninterrupted sitting conditions, interrupting sitting with resistance-type activity breaks had a significant effect on lowering postprandial glucose for both dietary conditions by ~ 36 mmol/L .

Conclusions: TV viewing, but not total volume or bouts of sedentary time, was positively associated with BMI and metabolic syndrome. Although some partial mediation effects were observed for *Study 1*, the combined findings from the four

studies showed dietary intake *did not* play a major role in the associations between the various sedentary behaviours and cardiometabolic risk factors examined. Further observational and experimental research is needed that assesses dietary intake concurrently with sedentary behaviour in order to confirm the role that dietary intake plays in the sedentary behaviour and cardiometabolic health relationship.

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Acknowledgement of authorship

I hereby certify that this thesis is in the form of a series of published papers of which I am a joint author. A list of co-authors contributing to the thesis is found below. I have included as part of the thesis, a written statement from each co-author attesting to my contribution to the joint publications. Each statement can be found in the Appendices. At the time of submission, five manuscripts in this thesis have been published in the following journals; *Obesity Reviews*, *Obesity Science & Practice*, *Obesity*, *BMC Public Health* and *Journal of Science and Medicine in Sport*. Where required, a copy of permission to include the manuscript in this thesis is found in the Appendix.

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Relevant to the thesis

1. **Fletcher EA**, Leech R, McNaughton SA, Dunstan DW, Lacy KE, Salmon J. Is the relationship between sedentary behaviour and cardiometabolic health in adolescents independent of dietary intake? A systematic review. *Obesity Reviews*. 2015;**16**(9): 795-805.
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Conference presentations

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List of abbreviations

The following abbreviations have been used in the current thesis. To note, there are discrepancies between the chapters that are not published (e.g. the Introduction and Discussion) and the chapters that have been published as manuscripts. Where possible, the Introduction and Discussion Chapters have avoided the use of the non-standard abbreviated terms, whereas the manuscripts have used the abbreviated versions that are outlined below:

BMI – Body mass index

BMI zscore – Body mass index z-score

CGM – Continuous Glucose Monitoring system

CPM – Counts per minute

FFQ – Food frequency questionnaire

HEI – Healthy Eating Index (score)

MetS – Metabolic syndrome

MVPA – Moderate-to-vigorous physical activity

RTLS – Real-time locating systems

SSB – Sugar-sweetened beverages

TV – Television (viewing)

List of definitions

Confounding factor –	a variable that influences both the dependent variable and independent variable causing a spurious association
Covariate –	a variable that is possibly predictive of the outcome under study
Mediator –	a variable that can explain the relationship between the dependent and the independent variable.
Sedentary behaviour –	any waking behaviours characterised by low energy expenditure (≤ 1.5 metabolic equivalents) while in a sitting or reclining posture.
Sedentary time –	overall time spent being sedentary is examined (e.g. by accelerometry)

SYNOPSIS

An abundance of evidence consistently shows sedentary behaviour is associated with an increased risk of obesity, metabolic syndrome, type 2 diabetes and cardiovascular disease in the adult population. However, less is understood about the cardiometabolic health effects of sedentary behaviour in the paediatric population. While it appears there is a consistent link between television (TV) viewing and obesity in children and adolescents, associations between obesity and other sedentary behaviours such as total volume of sedentary time and bouts of sedentary time are less clear. Given these mixed findings, other lifestyle behaviours, such as dietary intake, may contribute to the differences in associations observed. While numerous studies have reported consistent links between TV viewing and dietary intake, little is known about the role of dietary intake in the relationship between sedentary behaviour and cardiometabolic health outcomes.

Therefore, through a combination of literature reviews, secondary data analyses and original data collection, this multidisciplinary thesis-by-publication aims to improve our understanding of the role of dietary intake in the relationship between sedentary behaviour and cardiometabolic health outcomes in adolescents. It is important to note that, while the evidence in younger children (aged 5-12 years) is also unclear, adolescents aged 12-19 years have been specifically targeted for this thesis as various cardiometabolic risk markers are more likely to be established in this age group and track more strongly into adulthood as compared to younger children. This is partly due to the pubertal changes occurring during adolescence which can impact negatively on some cardiometabolic risk markers such as glucose and insulin. Furthermore, adolescence is also a time when major changes can occur in dietary

intake (e.g. an increase in energy-dense nutrient poor foods) and in sedentary behaviours (e.g. an increase in TV viewing time and decrease in physical activity).

The layout of this thesis by publication is as follows. In Chapter One, an overview of the literature relating to sedentary behaviour, dietary intake and cardiometabolic health in adolescents is provided. Chapter Two then provides a comprehensive systematic review of the literature examining whether sedentary behaviour is related to cardiometabolic health, independent of dietary intake. Chapter Three outlines the aims, objectives and hypotheses of the thesis. Chapters Four and Five examine the cross-sectional mediating role of dietary intake in the associations between TV viewing and cardiometabolic markers, and total volume and bouts of sedentary time with cardiometabolic markers, respectively. Chapter Six examines both the cross-sectional and prospective mediation effects of dietary intake on sedentary behaviour with body mass index (BMI). Chapter Seven uses an experimental design to examine the impact of uninterrupted sitting *versus* sitting with resistance-type activity breaks on adolescents' postprandial glucose responses following consumption of different meals varying in energy. Finally, Chapter Eight discusses the implications of the findings from Chapters Four to Seven in relation to the current literature and provides recommendations for future research.

CHAPTER ONE: INTRODUCTION

CHAPTER ONE: INTRODUCTION

1.1 Cardiometabolic risk factors in adolescents

Adolescents with chronic conditions, such as overweight and obesity, is a major public health concern in many Western countries.¹ For this thesis, overweight is defined as a body mass index (BMI) between the 85th and 95th percentile for adolescents of the same age and sex, and obesity is defined as a BMI at or above the 95th percentile for adolescents of the same age and sex.² Currently, rates of overweight and obesity have stabilised in Australia³ and other Western countries such as the United States (U.S.),⁴ Canada,⁵ Germany,⁶ and the Netherlands.⁷ However, globally, 22-24% of adolescents are overweight or obese, and 1 in 4 (27%) Australian adolescents are classified as overweight or obese³; thus overweight and obesity remains a major public health concern.⁸

Metabolic syndrome, a collection of risk factors (e.g. elevated waist circumference, blood pressure, triglycerides and glucose and/or low HDL-cholesterol) that increase a person's chance of developing type 2 diabetes and cardiovascular disease,⁹ is also a major public health concern.¹ Although the diagnostic criteria for metabolic syndrome in adolescents varies widely by country (see Appendix A), it is estimated that approximately 5.6% of adolescents worldwide have metabolic syndrome, with higher prevalence reported in overweight (10.7%) and obese (30.2%) adolescents.¹⁰ While no specific population-based prevalence of metabolic syndrome has been determined for Australian adolescents; findings from the Raine cohort (n = 1139, mean age 14 years) indicate that 28% of adolescents had a 'high risk score' for developing metabolic syndrome.¹¹ Since obesity tracks into adulthood,¹² and adolescents who have metabolic syndrome are at a higher risk of developing type 2 diabetes and

cardiovascular disease during adulthood,¹³ it is important to understand adolescent lifestyle risk factors to help inform effective interventions for the prevention of type 2 diabetes and future cardiovascular disease.

Lifestyle behaviours, such as physical inactivity and dietary intake, are well recognised risk factors for overweight and obesity¹⁴, and metabolic syndrome¹⁵ in adolescents. However, more recently, excessive sedentary behaviour – or *too much sitting* – has also emerged as a potential, independent risk factor for these chronic conditions.

1.2 Sedentary behaviour in adolescents

1.2.1 Introduction to sedentary behaviour

Over the past decade the term ‘sedentary behaviour’ has been used interchangeably with ‘physical inactivity’.¹⁶ However, strong observational evidence in adults shows that engaging in sedentary behaviour and being inactive are separate risk factors for cardiometabolic disease.^{17, 18} Additionally, it is possible for individuals who are meeting physical activity guidelines to also engage in high amounts of sedentary behaviour.^{19, 20} As a result, standardised definitions have recently been proposed by the Sedentary Behaviour Research Network²¹ (an international organisation that specifically focuses on the health impact of sedentary behaviour). The term *sedentary behaviour* is defined as “any waking behaviours characterised by low energy expenditure (≤ 1.5 metabolic equivalents) while in a sitting or reclining posture” (Figure 1) and the term *inactive* or *insufficiently active* refers to those who do not meet physical activity guidelines.

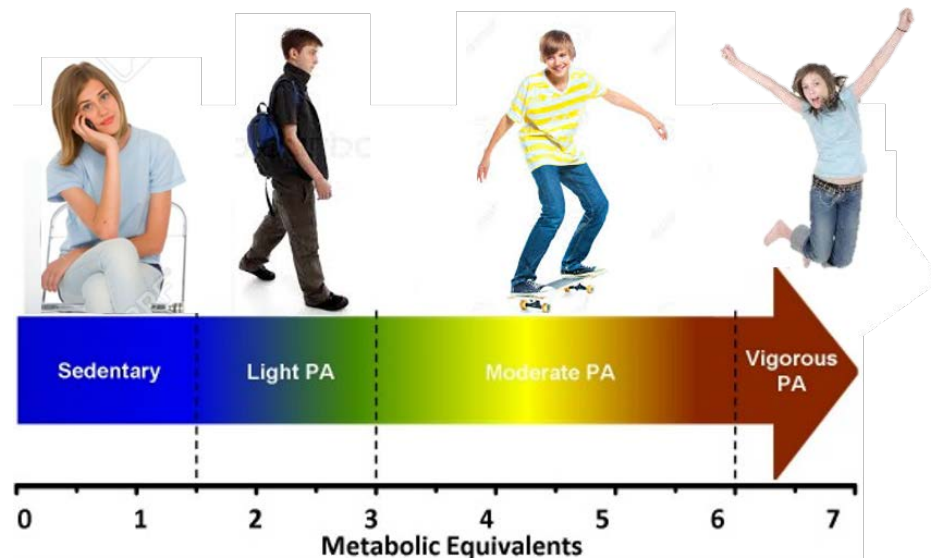


Figure 1: Definition of sedentary behaviour according to energy expenditure in metabolic equivalents (<http://www.sedentarybehaviour.org/>)²¹

There are also different nuances in the use of the term ‘sedentary’ depending on what aspect of sedentary behaviour is being assessed and how it is measured. For example, the term ‘sedentary behaviour’ is often used when the measure (usually via self-report) is able to identify participation in specific types of sedentary behaviours, such as TV viewing, computer use or playing sedentary video games. The term ‘sedentary time’ is mostly used when overall time spent being sedentary is examined (e.g. by accelerometry), and ‘sitting time’ is mostly used when time spent sitting is specifically examined either by self-report (e.g. how many hours do you spend sitting at work?) or when measured via inclinometers. In addition, the term ‘sedentary bouts’ is used to describe a period of time where sedentary time is uninterrupted. There are a number of ways to examine ‘sedentary bouts’. These include examining the average time spent in a sedentary bout, which is referred to as ‘average sedentary bout duration’, or examining short sedentary bouts (e.g. 5-minutes) versus prolonged

sedentary bouts (e.g. ≥ 45 -minutes). To ensure consistency, these sedentary terms will be used throughout the thesis.

1.2.2 Assessing sedentary behaviour

Various subjective (e.g. self-/proxy-report surveys, diaries) and objective (e.g. accelerometers, inclinometers, heart rate monitors, direct observations) methods are used to assess sedentary behaviour. When selecting a measure, key issues to be considered are the purpose or need, the validity and reliability, the appropriateness for use with specific populations, and the level of participant burden.²² The self-/proxy-report survey, accelerometer and inclinometer are the most commonly used measures in recent studies,²² therefore, these measures will be briefly discussed in reference to the adolescent population, with a summary of the key strengths and limitations of each method outlined in Table 1.

The self-/proxy-report survey is where the adolescent or their parent estimates the amount of time the adolescent spends in sedentary behaviours (e.g. how many hours do you [the adolescent], or your child watch TV?). Survey measures can provide rich data on different types of sedentary behaviours (TV viewing time, computer use, reading), whilst also providing an estimate of total sedentary time on a daily or weekly basis.²³ The self-/proxy-report survey measures are typically inexpensive to administer, have low participant burden, and are simple to analyse.²² However, as with most subjective measures, self-reporting has a number of limitations. First of all, self-/proxy-report surveys often collect data on only a single sedentary behaviour, such as TV viewing (e.g. “during the last 7 days, how much time [in hours or minutes per day] did you spend watching TV on a weekday / weekend day?”). As no single sedentary behaviour is representative of an individual’s total sedentary time,

this can only provide an estimate of sedentary levels. Secondly, surveys are retrospective, thus making them more susceptible to recall and reporting biases.²³ Additionally, due to advanced technologies, new types of sedentary behaviours are emerging, such as multi-tasking with screen devices, which adds to the complexity of measuring how much time is spent in screen-based and other sedentary behaviours.²⁴ Thus, in order to accurately assess average sitting time, multiple sedentary behaviours across different domains (e.g. TV viewing, reading, socialising, motorised travel) and time spent in various screen-based activities should be examined.²²

Conversely, an accelerometer, a small electronic device commonly worn on the hip via a belt, provides an objective measure of total sedentary time and time spent in sedentary bouts.²⁰ A hip-worn accelerometer records the volume, intensity and frequency of activity. The intensity is measured via the number of counts per minute (cpm), which can then be classified as either sedentary, light, moderate, or vigorous activity (e.g. the higher the cpm, the higher the intensity). Although the sedentary threshold has not been universally agreed upon, a threshold of 100 cpm has been consistently shown to have high sensitivity to assess sedentary time in an adolescent population.²⁵ One of the main advantages of using a hip-worn accelerometer is their accuracy to distinguish sedentary time in terms of low energy expenditure, as well as to identify the average time spent in sedentary bouts.²⁶ However, a key limitation of a hip-worn accelerometer is that they cannot determine posture (e.g. sitting *versus* standing still). This is due to the location of the device on the hip, making it difficult to capture postural changes and, consequently, periods of standing motionless may be misclassified as sitting. Accurately distinguishing between sitting and standing is important, as according to the Sedentary Behaviour Research Network definition,¹⁶

sedentary behaviour is when the person is in ‘a sitting position’ and it is this sitting position that is associated with adverse health outcomes.²⁰

Table 1: Strengths and limitations of a selection of common subjective and objective assessment methods of sedentary behaviour

	Overview	Strengths	Limitations
Self-/proxy-report survey	Adolescent (self) or their parent/guardian (proxy) estimates the amount of time they spend in sedentary behaviours	<ul style="list-style-type: none"> ➤ Provides rich data on different types of sedentary behaviours (TV viewing time, computer use, reading) ➤ Inexpensive ➤ Low participant burden ➤ Simple to analyse 	<ul style="list-style-type: none"> ➤ Susceptible to recall and reporting biases ➤ Often only collects data on a single sedentary behaviour ➤ Difficult to measure multi-tasking screen-based behaviours
Hip-worn Accelerometer	Small electronic device that records the volume, intensity and frequency of activity	<ul style="list-style-type: none"> ➤ Objective measure of average sedentary time based on low accelerometer counts below a specified threshold ➤ Objective measure of bouts of sedentary time 	<ul style="list-style-type: none"> ➤ Cannot distinguish between sitting <i>versus</i> standing still ➤ Lacks context specific information on the types of sedentary behaviours
Thigh-worn Inclinometer	Small device worn on the front of the thigh that has the ability to detect postural changes e.g. sitting, lying or standing	<ul style="list-style-type: none"> ➤ Objective measure of sitting or lying time 	<ul style="list-style-type: none"> ➤ Lacks context specific information on the types of sedentary behaviour

Alternatively, an inclinometer, a small device commonly worn on the front of the thigh, has the ability to detect postural changes, such as whether someone is sitting, lying or standing.²⁷ As such, thigh-worn inclinometers are becoming a more common

measure in studies involving youth to determine how much of their day is actually spent sitting. Although accelerometers and inclinometers provide an objective measure of sedentary and sitting time, they both lack information on the *type* of sedentary behaviour, such as watching TV or using the computer. This is important, as new evidence shows that various types of sedentary behaviours, such as TV viewing, may have different relationships with cardiometabolic health as opposed to other sedentary behaviours.²⁸ Thus, researchers are strongly encouraged to use a combination of both objective and subjective measures (e.g. accelerometers and survey) to ensure accurate reporting of sedentary time whilst gathering information on the different types of sedentary behaviours in which participants have engaged.²³ This information can then be used to accurately report on the prevalence and trends of specific population groups engaging in sedentary behaviours.

1.2.3 Sedentary behaviour recommendations and prevalence

Currently, the Australian Physical Activity and Sedentary Behaviour guidelines recommend that children aged 5-17 years should limit their use of electronic media for entertainment to less than 2 hours per day and break up long periods of sitting as often as possible.²⁹ These guidelines also emphasise the importance of sitting less, and encourage those already meeting the physical activity guidelines to still follow these recommendations for optimal health. These guidelines are similar to the recommendations from professional societies like the American Academy of Pediatrics Committee³⁰ and the Canadian Society for Exercise Physiology,³¹ where it is recommended that children and adolescents engage in no more than 2 hours of screen time per day.

Despite these recommendations, less than one third of Australian youth aged 5-17 years are meeting the screen time guidelines.³² This is similar to other developed countries, where 27% of U.S. adolescents aged 12-15 years³³ and 49% of Canadian adolescents aged 12-18 years³⁴ engage in more than 2 hours of screen time per day. Additionally, in Australia, adolescents both exceed the screen time recommendations and spend nearly half of their daily leisure time (approximately 4.6 to 6.6 hours/day) being sedentary.³⁵ This is similar to findings from the International Children's Accelerometry Database (n = 20,871 from the U.S., Brazil, Europe), which showed that youth aged 4-18 years on average spent 5.9 waking hours/day being sedentary.³⁶ As the majority of adolescents in many Western countries are exceeding the screen time recommendations and spending nearly half of their leisure time being sedentary, it is important to understand the adverse effects of *too much* sedentary behaviour has on cardiometabolic health. Thus, the next section will outline the various associations between sedentary behaviour and cardiometabolic risk markers in adolescents.

1.2.4 Associations between sedentary behaviour and cardiometabolic health

Television viewing and cardiometabolic health

There is an abundance of epidemiological evidence to show a consistent and positive relationship between TV viewing and an increased risk of overweight and obesity among adolescents.^{28, 37, 38} In 2011, a systematic review on the relationship between sedentary behaviour and health indicators in youth aged 5-17 years reported 94 of 119 (79%) cross-sectional studies observed positive associations between TV viewing and markers of adiposity.³⁷ This systematic review was updated in early 2016 (with additional searches performed to find relevant new studies published after 2010) and found similar consistent findings, with 58 out of the 71 cross-sectional studies (82%)

published after 2010 reporting a positive association between TV viewing and overweight and obesity.²⁸ Additionally, the intervention studies (n = 10) and longitudinal studies (n = 35) included in both systematic reviews revealed a dose-response relationship; those who watched more TV were more likely to be overweight or obese.^{28, 37}

In contrast to the adiposity literature, few studies have found positive associations between TV viewing and other cardiometabolic markers in adolescents such as insulin, glucose, blood pressure, and cholesterol. For example, some studies have reported positive associations between TV viewing and fasting insulin, HOMA-IR,³⁹ insulin resistance,⁴⁰ glucose,⁴¹ hypertension,⁴² and reduced HDL cholesterol^{41, 43} in adolescents, whilst other studies have reported either few or null findings with individual cardiometabolic markers.^{44, 45} There is also mixed evidence on associations between TV viewing and a combined cardiometabolic risk score or metabolic syndrome in adolescents. Whilst the earlier systematic review from 2011 found that all eleven studies (two longitudinal, nine cross-sectional) that had assessed TV viewing found positive associations with an increased risk for metabolic syndrome or cardiovascular risk score,³⁷ the updated systematic review reported mixed findings from the twelve new studies (two longitudinal, ten cross-sectional) included.²⁸

Sedentary time and cardiometabolic health

Despite the consistent evidence between TV viewing and overweight and obesity, *total volume* of sedentary time appears to be inconsistently associated with markers of adiposity in the adolescent population. The updated systematic review reported that only three out of nine longitudinal studies and 3 out of 18 cross-sectional studies found significant positive associations between accelerometer-measured total volume

of sedentary time and overweight and obesity.²⁸ However, when examining sedentary bouts with markers of adiposity, more consistent associations have been observed. Some cross-sectional studies, but not all,⁴⁴ have found that the number of sedentary bouts lasting > 10 minutes was positively associated with BMI⁴⁶ and breaking up sedentary time was associated with a lower waist circumference.²⁶

When examining other cardiometabolic health outcomes such as metabolic syndrome and individual cardiometabolic risk markers, few studies have reported significant associations with total volume of sedentary time and bouts of sedentary time. For example, the 2016 systematic review reported that only one of four studies found positive associations between total volume of sedentary time and metabolic syndrome.²⁸ Similarly, when examining individual cardiometabolic risk markers, the 2016 systematic review reported that only one of six studies found a significant inverse association between total volume of sedentary time and HDL cholesterol. Whereas, other studies consistently reported null associations between total volume and bouts of sedentary time with blood pressure, insulin and glucose.²⁸

In summary, depending on the type of sedentary behaviour being examined, there appears to be mixed associations between sedentary behaviour and cardiometabolic risk markers in the adolescent population. For example, specific sedentary behaviours, like TV viewing, appear to be consistently and positively related to markers of adiposity and some cardiometabolic risk markers; whereas very few studies have found significant associations between total volume and bouts of sedentary time and markers of cardiometabolic health. Given these differences, section 1.2.4 will explore the potential behavioural mechanisms that may explain why stronger associations are often observed for TV viewing and cardiometabolic health compared to total volume or bouts of sedentary time. However, before the

behavioural mechanisms are explored, it is important to firstly understand the physiological mechanisms that have been proposed that could explain why sedentary behaviour is linked with adverse health.

1.2.5 Physiological mechanisms of sedentary behaviour and cardiometabolic health

There are two key physiological mechanisms proposed to explain the adverse health associations related to sedentary behaviour. The first proposed mechanism relates to the physiology of muscle contractile activity. For example, uninterrupted sitting leads to a reduction in muscle contractile activity (particularly of the lower limbs) which is associated with reduced blood flow and reduced efficiency of many of the body's regulatory processes, including the transport of blood glucose from circulation into the muscle.¹⁷ High amounts of glucose in the blood (e.g. $\geq 5.6\text{mmol/L}$) is detrimental to health as it causes long-term damage to the blood vessels that supply vital organs. Thus, high blood glucose is a well-known risk factor for cardiometabolic diseases such as obesity and type 2 diabetes.⁹ The second physiological mechanism proposed is that engaging in sedentary behaviour may simply reduce the overall metabolic rate,⁴⁷ and thus contribute to weight gain and consequent cardiometabolic disease.

On the other hand, breaking up prolonged periods of sedentary time has been shown to have beneficial outcomes on cardiometabolic risk markers in the adult population, including reduced waist circumference, BMI, triglycerides, postprandial glucose,⁴⁸ insulin levels,⁴⁹ and C-peptide levels.⁵⁰ Similar findings have also been observed in adults with type 2 diabetes,⁵¹ young adults (aged 18-24 years),⁵⁰ and recently in children (aged 7-11 years).⁵² The suggested mechanisms underlying these findings

are thought to be due to the opposite of those described above; that is, an increase in skeletal muscle contractions which facilitates glucose uptake, or frequent breaks in sitting time contributing to a higher total energy expenditure and prevention of weight gain.⁴⁸

1.2.6 Behavioural mechanisms of sedentary behaviour and cardiometabolic health

Three key behavioural mechanisms have been proposed to explain why TV viewing, but not total volume of sedentary time, is consistently linked with cardiometabolic health: 1) the adverse biological effects of the TV screen itself;⁵³⁻⁵⁵ 2) the physical activity displacement hypothesis;^{56, 57} and 3) an increase in dietary intake.⁵⁸ Although further research is required, the biological effects of the TV screen itself (e.g. the blue light emitted from the TV screen) is thought to suppress the release of melatonin and reduce sleep quality at night^{53, 54}. Insufficient sleep can then increase the risk of overweight and obesity⁵⁵, as well as impact negatively on certain cardiometabolic risk markers such as blood pressure and insulin.⁵⁴

Secondly, some evidence suggests that time spent watching TV displaces overall physical activity, leading to a reduced energy expenditure and, thus, an increased risk of adverse cardiometabolic health.^{56, 57} However, in a recent meta-analysis involving 163 studies on associations between sedentary behaviour and physical activity in children and adolescents, only a small, negative association between TV viewing and physical activity ($r = -0.06$, 95% CI -0.08 to -0.05) was found.⁵⁹ Although the direction of the association supports the displacement hypothesis, the small effect size implies that TV viewing and physical activity are not strongly related, and that these behaviours may co-exist.

The third potential behavioural mechanism that could explain why stronger associations are observed for TV viewing and cardiometabolic health is through changes in dietary intake or the displacement of healthy eating behaviours. This is thought to be due to TV viewing being consistently linked with a high consumption of discretionary foods and sugar-sweetened beverages, and a low consumption of fruit and vegetables;⁵⁸ behaviours that contribute to weight gain and to other adverse cardiometabolic outcomes. However, when specifically exploring the mediation effects of dietary intake on the association between TV viewing and cardiometabolic health, the evidence is mixed. Studies in pre-school age children⁶⁰ and adults⁶¹ have reported that the association between TV viewing and obesity was partially mediated by the consumption of food and beverages, whilst another study in adolescents have reported no mediating effects.⁶²

Therefore, although TV viewing has been consistently linked with unhealthy dietary intake, the role of dietary intake in the TV viewing and cardiometabolic health relationship is unclear. Furthermore, it is also unclear whether dietary intake could also explain the mixed findings observed between other types of sedentary behaviours and cardiometabolic health outcomes. For example, it is possible that the mixed findings observed between total volume and bouts of sedentary time with cardiometabolic health may be due to some studies controlling for dietary intake in the analyses whilst other studies do not. Before these issues can be explored, it is important to provide an overview of concepts related to dietary intake that are relevant to this thesis.

1.3 Dietary intake in adolescents

1.3.1 Introduction to dietary intake

Dietary intake is a multifaceted lifestyle behaviour. For this thesis, ‘dietary intake’ will be used as the umbrella term to define all food and beverages consumed.

However, the term ‘dietary intake’ can also refer to ‘nutrient intakes’, ‘meal patterns’ and ‘dietary patterns’. Given the large scope of the dietary literature in adolescents, this thesis will primarily focus on the ‘foods and beverages consumed’, with particular emphases on the foods and beverages related to either sedentary behaviour or cardiometabolic risk markers.

1.3.2 Assessing dietary intake

Assessing dietary intake is complex. Currently, there is no one method that can accurately measure all of the different foods and beverages an individual consumes, and the various combinations and patterns in which foods are consumed. There are various methods of recording dietary intake in adolescents, with the most common being the 24-hour dietary recall (interviewer-administered), food frequency questionnaire and food record.⁶³ These methods can be completed by adolescents or via proxy report (e.g. parent or guardian completes it on behalf of their child). As a guideline, it is recommended that proxy reports are used for children younger than 12 years of age, due to children’s lower literacy levels and limited cognitive abilities to recall their own dietary intake.⁶⁴ As the 24-hour dietary recall, food frequency questionnaire and food record are the most commonly used in adolescent studies, these methods will be briefly discussed, with an overview of the strengths and limitations of each method provided in Table 2.

Table 2: Strengths and limitations of a selection of common assessment methods of dietary intake

	Overview	Strengths	Limitations
24-hour dietary recall (Interviewer-administered)	Interviewer asks participant to list all foods and beverages consumed on the previous day.	<ul style="list-style-type: none"> ➤ Considered the ‘gold-standard’ measure ➤ Measures the quantity of food and beverages consumed ➤ Open-ended questions allows comprehensive and detailed report of the preparation and cooking methods used ➤ Detailed information can be captured (e.g. time of day, source of food and portion sizes) ➤ Individuals cannot change their eating behaviour 	<ul style="list-style-type: none"> ➤ Multiple days required to capture usual intake ➤ Susceptible to social desirability bias ➤ Susceptible to recall error
Food frequency questionnaire	Captures retrospective dietary intake by participant completing a questionnaire	<ul style="list-style-type: none"> ➤ Provides information on the usual consumption of foods and beverages over a longer time period (e.g. over the last month or year) 	<ul style="list-style-type: none"> ➤ Susceptible to social desirability bias ➤ Susceptible to recall error
Food record/diary	Participant records all foods and beverages consumed at the time of consumption over a specified period	<ul style="list-style-type: none"> ➤ Measures the quantity of food and beverages consumed 	<ul style="list-style-type: none"> ➤ High participant burden ➤ Individuals can change their eating behaviour ➤ Susceptible to social desirability bias

The 24-hour dietary recall is considered to be the ‘gold-standard’ measure and involves the participant listing what foods and beverages they consumed on the previous day. Traditionally, the participant completes the 24-hour dietary recall with an interviewer, however, automated 24-hour recalls have recently been established in

Australia⁶⁵ and the U.S.⁶⁶ where participants complete the 24-hour dietary recall online. Overall strengths of the interviewer-administered 24-hour dietary recall include assessing the quantity of food and beverages consumed, detailing the preparation and cooking methods used as well as information about the time of day the food/beverage was consumed and portion sizes. Further, individuals cannot change their eating behaviour; a common problem often reported when using food records due to individuals recording their food at the time of consumption.⁶³

However, a major limitation of the 24-hour dietary recall is that multiple days of recall are required to get an accurate reflection of usual intake. Additionally, social desirability bias is common whereby individuals may intentionally underreport foods and beverages consumed. Alternatively, individuals may unintentionally forget certain foods and beverages or inaccurately report portion sizes consumed.⁶³

An alternative dietary assessment method that also captures retrospective dietary intake is a food frequency questionnaire. These questionnaires are often standardised and provide information on the usual consumption of foods and beverages consumed over a longer time period (e.g. over the last month or year).⁶⁷ Some food frequency questionnaires also report on serving sizes, however, similarly to the 24-hour dietary recall, self-reported serving sizes are susceptible to inaccurate reporting. Another limitation of a food frequency questionnaire is that individuals can misrepresent the frequency with which foods and beverages are consumed.⁶³

Food records, also known as food diaries, provide another way to collect dietary intake data. However, unlike the 24-hour dietary recall and food frequency questionnaire, the foods and beverages consumed are generally recorded at the time of consumption over a specified period.⁶⁸ It is suggested that three days is a sufficient

period of time to estimate regular food intake without being too labour intensive for the participant.⁶⁸ Food records can also include weighing foods and beverages. However, this adds to the high participant burden, and consequently may lead to participants changing their diet so it is easier to record. An alternative is using household measures to determine food and beverage volumes and/or taking photos of foods, beverages or meals before consuming them.⁶⁸ Taking photos is a relatively new method aimed at reducing participant burden, however, further research is required to fully understand if this is the case.

1.3.3 Dietary recommendations and intake prevalence

The Australian Dietary Guidelines released in 2013 provides up-to-date recommendations about the amount and types of foods children and adults should eat for optimal health and wellbeing.⁶⁹ Similar to previous guidelines, the dietary guidelines for adolescents aged 12-18 years are divided into two categories: foods to eat every day and foods to limit. Foods to eat everyday include a wide variety of nutritious foods from the five food groups: 1) vegetables and legumes; 2) fruit; 3) wholegrain foods such as breads, rice and oats; 4) lean meats such as red meat, poultry and fish and/or their alternatives such as eggs, nuts and seeds; and 5) dairy foods such as milk, yoghurt, cheese and/or their alternatives. For information on the specific recommended serves and serving sizes for Australian adolescents according to age and gender, please see Table 3.

Foods to limit are referred to as ‘discretionary foods’ as they are not an essential part of the diet. These foods often contain high levels of saturated fat, added salt and added sugars. Examples of discretionary foods with high levels of saturated fat and added salt include biscuits, cakes, pastries, pies, processed meats, commercial

Table 3: Recommended serving sizes for Australian adolescents aged 12 to 18 years according to the Australian Dietary Guidelines 2013⁶⁹

	Serves per day		Examples of serving sizes
	12-13 years	14-18 years	
Vegetables & legumes			
Boys	5.5	5.5	½ cup of vegetables, 1 medium potato, 1 cup of salad
Girls	5	5	
Fruit			
Boys	2	2	1 apple, 1 banana, 2 small Apricots
Girls	2	2	
Grain foods			
Boys	6	7	1 slice bread, ½ cup rice, ½ cup cooked pasta
Girls	5	7	
Lean meats/alternatives			
Boys	2.5	2.5	80g chicken, 1 tin of tuna, 2 eggs
Girls	2.5	2.5	
Dairy/alternatives			
Boys	3.5	3.5	1 cup milk, 2 slices cheese, ¾ cup yoghurt
Girls	3.5	3.5	

burgers, pizza, fried foods, potato chips, crisps, and other savoury snacks. Foods and drinks containing added sugars include confectionary, sugar-sweetened soft drinks and cordials, fruit drinks, vitamin waters and energy and sport drinks.⁶⁹

When dietary intakes are compared with the Guidelines, adolescents in Australia have one of the worst diets among the Australian population.⁷⁰ Results from the 2011-2013 Australian Health Survey found only 5% of adolescents consumed enough fruit and vegetables and over 50% of adolescents consumed sugar-sweetened beverages on the day the survey was completed.⁷⁰ Similar results are also observed in the U.S. and Europe, where it is reported that adolescents are consuming more sugar-sweetened beverages and fast food than any other age group.^{71, 72} These results are concerning, as a diet low in vegetables and fruits and high in discretionary foods and sugar-sweetened beverages, increases the risk of developing chronic conditions such as obesity, type 2 diabetes and cardiovascular diseases during adulthood.^{73, 74}

1.3.4 Associations between dietary intake and cardiometabolic health

Previous evidence in adults has consistently found that an energy-dense diet that is low in fruits, vegetables and dietary fibre is associated with an increased risk of obesity, type 2 diabetes and cardiovascular disease.^{73, 74} However, less is understood about associations between various dietary elements and cardiometabolic health risk markers in adolescents. High intakes of discretionary foods (e.g. salty snacks, sweets and desserts)⁷⁵ and sugar-sweetened beverages,⁷⁶⁻⁷⁹ and low intakes of fruit and vegetables⁸⁰ have been associated with an increased risk of obesity. However, there is very little evidence available specifically in adolescents examining the associations between dietary intake with other cardiometabolic risk markers such as glucose, insulin and blood pressure or with other chronic conditions such as metabolic syndrome.

Of the available evidence examining individual cardiometabolic risk markers, cross-sectional studies have found associations with fruit and vegetable intake and sugar-sweetened beverage consumption. For example, a cross-sectional study in Mexican youth aged 9-13 years found associations between low fruit and vegetable intake and high sugar-sweetened beverage consumption with increased plasma glucose and diastolic blood pressure.⁸¹ Similarly, cross-sectional studies in Australian adolescents have found a diet high in fruit, salad, cereals and fish was inversely associated with diastolic blood pressure,⁸² and a diet high in vegetarian foods was associated with favourable waist circumference, total cholesterol/HDL ratio and LDL cholesterol.⁷⁶

As opposed to individual foods or beverages, research on the overall quality of the diet has yielded relatively consistent findings with respect to metabolic syndrome in adolescents. Data from the 1999-2002 National Health and Nutrition Examination

Survey (NHANES) adolescent cohort (aged 12-19 years) showed that a higher overall Healthy Eating Index (HEI) score and fruit score (a component of the HEI) were associated with a lower prevalence of metabolic syndrome, even after adjusting for BMI and physical activity intensity.⁸³ In addition, results from 1139 Australian adolescents (aged 14 years) participating in The Raine Study reported a 'Western diet' consisting of a high intake of takeaway foods, refined grains, confectionary and soft drinks was associated with greater odds for metabolic syndrome (OR = 2.50; 95% CI 1.05 to 5.98) in girls, but not in boys.¹¹ These cross-sectional findings suggest that the overall quality of the diet may also be important for optimal cardiometabolic health. Therefore, in addition to the thesis examining 'food and beverage intakes', overall dietary quality will also be explored.

As previously proposed, dietary intake may also play an important role in the link between sedentary behaviour and cardiometabolic health. As such, the next section will explore the associations between dietary intake with various 'types' of sedentary behaviour (i.e. TV viewing, total sedentary time, bouts of sedentary time, and prolonged sedentary time), specifically in the adolescent population.

1.3.5 Associations between sedentary behaviour and dietary intake

Screen time and dietary intake

There is an abundance of evidence on the associations between TV viewing and elements of a less healthy diet in adolescents^{58, 84}. In a review by Pearson et al.,⁵⁸ of 43 studies examining the associations between sedentary behaviour and dietary intake in children and adolescents, TV viewing was consistently found to have positive associations with total energy intake, the consumption of discretionary foods, sugar-sweetened beverages, and fast food, and inverse associations with fruit

and vegetable consumption. Similarly, a large Australian cross-sectional study (n = 18,486) found that secondary students (aged 12-17 years) who watched TV ≥ 3 h/day were more likely to consume fast food meals, discretionary foods and sugar-sweetened beverages when compared with students who watched TV ≤ 2 hours/day; even after controlling for physical activity, socioeconomic status and other demographic variables.⁸⁵ Further, data collected from a large cross-national study involving Central and Eastern European countries⁸⁶ and data collected from large national studies from the U.S.^{87, 88} and Canada⁸⁸ also found that adolescents who watched more TV^{86, 87} or had a TV in the bedroom⁸⁸ were more likely to consume fast food, sweets and soft drinks on a daily basis, and less likely to consume fruit and vegetables.

Experimental studies in adolescents examining food consumption while engaging in screen-based behaviours (e.g. TV viewing, video game playing, and computer use) have also found consistent associations with adolescents' energy intake.^{89, 90} For example, in a randomised, cross-over study by Chaput et al.,⁸⁹ 22 healthy male adolescents (aged 15-19 years) underwent two 1-hour experimental conditions (video game playing and sitting only), followed by an *ad libitum* lunch and questionnaire on appetite. After a single session of video game playing, adolescent males increased their food intake by 335 kilojoules, regardless of their perceived appetite. Further, in another controlled experimental study by Epstein et al.,⁹⁰ 16 healthy adolescents (aged 12-16 years) either increased their screen-based sedentary behaviours (i.e. TV viewing, video game playing and computer use) by 25-50% over a 3-week period or decreased their sedentary behaviours by 25-50% over a 3-week period. Findings showed that when screen-based sedentary behaviours were decreased by 100 min/day, total energy intake and fat intake also decreased by 17% and 32%,

respectively, with no significant changes in energy intake when sedentary behaviours were increased. The authors suggested that the reduction in fat intake may be due to the participants decreasing their consumption of high-fat snack foods, which are often consumed while watching TV or during other sedentary behaviours.

Sedentary time and dietary intake

In contrast to the literature on screen-based behaviours and dietary intake, associations between total volume of sedentary time or bouts of sedentary time and dietary intake in adolescents have been less frequently studied. In fact, no observational study to date has examined associations between accelerometry-measured overall sedentary time or bouts of sedentary time and dietary intake specifically in adolescents. However, there is some evidence available in younger children. For example, in a cross-sectional study examining the differences between TV viewing and total volume of sedentary time with dietary intake in Canadian children (aged 9-11 years),⁹¹ the authors found that TV viewing was strongly associated with a number of unhealthy dietary behaviours, such as the consumption of sweets, pastries, potato chips, fried foods, fast foods and ice-cream, independent of sedentary time and other covariates. However, total sedentary time was only found to be negatively associated with the consumption of sports drinks, independent of TV viewing time.

In contrast, findings from the SPEEDY (Sport, Physical activity and Eating behaviour: Environmental Determinants in Young people) cohort involving 1,317 children (aged 9-10 years) from the United Kingdom reported associations between total volume of sedentary time and soft drink consumption and savoury snacking.⁹² However, the association was not in the expected direction, with boys that were in

the highest quartile for soft drink consumption and savoury snack consumption spending 14 and 12 minutes *less* in sedentary time each day, respectively. These findings suggest that for children, TV viewing time may be more strongly associated with unhealthy dietary behaviours than total sedentary time. However, due to the lack of evidence available in adolescents, further research is required to investigate whether similar associations exist in an adolescent population.

Further, findings from experimental studies examining prolonged sitting and *ad libitum* food intake have reported no significant differences in food intake when sitting was prolonged or when sitting was interrupted with exercise.^{52, 93} In a randomised study by Saunders et al.,⁹³ 20 healthy adolescents (aged 10-14 years) participated in the following three conditions on separate days: uninterrupted sitting; a day of sitting with light-intensity breaks; and a day of sitting with light- and moderate-intensity breaks, followed by a buffet meal in the afternoon. Findings showed no significant differences in *ad libitum* food intake immediately following the three experimental conditions ($p < 0.25$). In a similar study design involving uninterrupted sitting and sitting with moderate-intensity walking breaks in children (aged 7-11 years), Belcher and colleagues⁵² found that total energy intake and the percentage of kilojoules consumed from carbohydrates, protein and fat did not significantly differ between the two experimental conditions. These findings suggest that children *do not* compensate by reducing their food intake when they are exposed to a long bout of sedentary behaviour.

1.4 Summary

Overall, there appears to be consistent evidence of the relationship between TV viewing and obesity, and some evidence to show links between TV viewing with

other cardiometabolic risk markers such as insulin resistance, hypertension and reduced HDL cholesterol in the adolescent population. It is also well established that TV viewing is strongly associated with unhealthy dietary behaviours such as a higher energy intake, a high consumption of discretionary snacks and sugar-sweetened beverages, fast food intake and low fruit and vegetable consumption. However, due to the lack of research, what is less clear, is the evidence involving total volume of sedentary time and bouts of sedentary time, both in regards to associations with cardiometabolic risk markers and dietary intake.

The mixed findings between TV viewing and total volume of sedentary time suggest that there may be other factors involved that partially explain the stronger relationships found between TV viewing and cardiometabolic health. Given that TV viewing has been shown to have consistent associations with dietary intake, it is worth exploring its role in the relationship between TV viewing and cardiometabolic health. However, little is known about whether dietary intake plays a key role when total volume or bouts of sedentary time are examined. As such, a systematic review of the literature was undertaken by the candidate to summarise the associations between various sedentary behaviours and cardiometabolic risk markers in adolescents, while either accounting for dietary intake in the analyses or specifically conducting mediation analyses. The findings from the systematic review are published in *Obesity Reviews* and is presented in the next Chapter.

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CHAPTER TWO: SYSTEMATIC REVIEW

**SYSTEMATIC REVIEW OF SEDENTARY
BEHAVIOUR, DIET AND CARDIOMETABOLIC
HEALTH IN ADOLESCENTS**

CHAPTER TWO: SYSTEMATIC REVIEW

2.1 Overview

As previously discussed in Chapter One, the role dietary intake has in the relationship between various sedentary behaviours and cardiometabolic health outcomes in adolescents is unclear. Therefore, a systematic review was conducted by the candidate to identify all studies that have either performed mediation analyses or accounted for dietary intake when examining associations between sedentary behaviour and cardiometabolic risk factors. The findings from the systematic review will be used to identify the gaps within the sedentary behaviour literature on adolescents and guide the objectives of the studies included in this thesis.

The following systematic review has been published as a manuscript in *Obesity Reviews* (Impact Factor: 7.995) and has been formatted according to their requirements. The review was prospectively registered with PROSPERO; CRD42014010359. The citation for the review is: *Fletcher E, Leech R, McNaughton SA, Dunstan DW, Lacy K, Salmon J. Is the relationship between sedentary behaviour and cardiometabolic health in adolescents independent of dietary intake? A systematic review. Obesity Reviews. 2015;16(9): 795-805.* The manuscript supplementary files can be found directly after the manuscript in *Section 2.3*. The ‘Authorship Statement’ for this manuscript and a copy of the ‘Permission Statement to Publish’ this manuscript are contained in Appendix B.1 and Appendix B.2, respectively.

Findings from this review were presented at the International Society of Behavioral Nutrition and Physical Activity conference in Edinburgh, Scotland in June 2015 and at the 10th Research Degree Symposium held by the School of Exercise and Nutrition Sciences, Deakin University in Melbourne, Australia in September 2014 (abstracts available in Appendix B.3 and Appendix B.4, respectively).

The author contributions to this review were as follows, with the candidate contributing to all aspects of the manuscript process: Fletcher, McNaughton, Dunstan, Lacy and Salmon designed the review; Fletcher and Leech reviewed all articles based on the inclusion and exclusion criteria and performed the methodological quality assessment of the included studies. Fletcher performed the data extraction of all included studies. Fletcher wrote the initial draft of the manuscript, whilst Leech, McNaughton, Dunstan, Lacy and Salmon provided critical edits and editions. All authors approved the final manuscript.

2.2 Manuscript

Obesity Comorbidity

Is the relationship between sedentary behaviour and cardiometabolic health in adolescents independent of dietary intake? A systematic review

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Summary

Screen time, but not overall sedentary behaviour, is consistently related to cardiometabolic health in adolescents. Because of the associations screen time has with dietary intake, diet may be an important factor in the screen time and health relationship; however, evidence has not previously been synthesized. Thus, the aim of this systematic review was to explore whether the associations between various sedentary behaviours and cardiometabolic risk markers are independent of dietary intake in adolescents. Online databases and personal libraries were searched for peer-reviewed original research articles published in English before March 2014. Included studies assessed associations between sedentary behaviour and cardiometabolic markers in 12- to 18-year-olds and adjusted for dietary intake. Twenty-five studies met the inclusion criteria. From the 21 studies examining sedentary behaviour and adiposity, the majority found significant positive associations between television viewing, screen time and self-reported overall sedentary behaviour with markers of adiposity, independent of dietary intake. No significant associations between screen time with blood pressure and cholesterol were reported. Sedentary behaviour appears to be associated with adiposity in adolescents, irrespective of dietary intake. However, the variability of dietary variables between studies suggests further work is needed to understand the role of dietary intake when examining these associations in youth.

Keywords: Adolescents, cardiometabolic, diet, screen time.

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Introduction

Cardiometabolic risk factors, specifically obesity, insulin resistance, hypertension and dyslipidaemia, are becoming more prevalent among adolescents (1,2). In Western countries, 25–33% of adolescents are overweight or obese (3–5) and 5–10% of adolescents have one or more of the other cardiometabolic markers (1). Spending large amounts of time using screen-based media, in particular time spent watching television, has been consistently associated with cardiometabolic health in children and adolescents (6–10). This is concerning since current figures indicate that

62–83% of adolescents from Westernized countries are exceeding the screen-based recommendations of no more than 2 h of screen time per day (5,11,12).

In contrast to the consistent links shown for television viewing and screen time, there is inconsistent evidence of associations between overall sedentary time (usually measured by accelerometry) and cardiometabolic risk markers in youth (13,14). This inconsistency suggests that some of the observed associations between screen time and health may be explained by other factors, rather than time spent sedentary or 'sitting' *per se*. There is a possibility that the differences may be simply due to different measures used,

with self-reported measures and objective measures reported to have low to moderate correlations (−0.71 to 0.96) (15).

Alternatively, there may be other lifestyle behaviours such as physical activity and dietary intake that may explain the stronger relationship seen between screen time and health. For example, there is some evidence indicating that adolescents who spend more time in screen time behaviours are less physically active (16,17). However, findings from a recent systematic review reveal screen time and physical activity among youth are not strongly inversely associated ($r = 0.080$, 95% CI −0.101, −0.060) (18), suggesting they are independent behaviours. Additionally, most studies that report associations between screen time and health outcomes in this age group account for physical activity levels (9,19).

In youth, there is consistent evidence that television viewing is linked to elements of a less healthy diet such as lower fruit and vegetable consumption, higher consumption of energy-dense snacks, drinks, and fast foods, and higher total energy intake (20). This suggests that dietary intake may play a role in the relationship between television viewing and cardiometabolic health. However, the mediating role of dietary intake has rarely been examined in youth, while the few studies among adults have found no consistent relationship (21–24). There is now an emerging amount of literature that has examined the relationship between sedentary behaviour and cardiometabolic risk markers in adolescents that has accounted for dietary intake in the analyses. However, there have been no systematic reviews to determine whether these associations are consistent, independent of dietary intake, or whether the studies are of sufficiently high quality. With the majority of Western adolescents not meeting the current screen time guidelines (5,11,12) and the increase in prevalence of cardiometabolic conditions such as obesity, type 2 diabetes and cardiovascular disease (1,2), adolescence is an important time to encourage healthy behaviour changes. Therefore, the aim of this systematic review was to identify and synthesize evidence from studies that have accounted for dietary intake when examining the relationship between sedentary behaviour and cardiometabolic health in 12- to 18-year-old adolescents.

Methods

This review is registered with PROSPERO, CRD42014010359.

Search strategy

Online databases (Medline, Global Health, PsycInfo, Web of Science and Embase), reference lists and personal libraries were searched for peer-reviewed original research

articles published in English before 25 March 2014. The following keyword combinations were used for *sedentary behaviour* (sedentar*, sitting, indoor*, screen time, computer*, television, inactivit*, video game*, internet), *dietary intake* (diet*, nutrition, food*, snack*, drink*, beverage*, eating, energy intake*, meals), *health outcome* (overweight, obes*, adiposity, waist circumference*, waist-hip ratio, body mass index, blood pressure, hypertens*, glucose intolerance, blood glucose, insulin, cholesterol, lipoprotein, triglycerides, lipid metabolism, cardiometabolic, metabolic syndrome, cardiovascular disease*) and *age* (youth, adolescen*, child*, young people). The search terms were restricted to title and abstract only.

Inclusion and exclusion criteria

To be included in the review, studies were required to meet the following criteria: (i) be published as a peer-reviewed original research article, with full-text availability; (ii) the study participants' mean age was between 12 and 18 years, or for longitudinal studies the mean age was between 12 and 18 years at baseline or at follow-up; (iii) the study included a measure of sedentary behaviour as an independent variable defined by 'any waking behaviours characterized by low energy expenditure (≤ 1.5 METS) while in a sitting or reclining posture' (25), and not the absence of sufficient levels of physical activity; (iv) the statistical analyses included an adjustment for dietary intake as defined by the intakes of energy, macronutrients, foods, and/or beverages) and (v) the study assessed at least one cardiometabolic risk marker as the main outcome (i.e. adiposity, blood pressure; insulin sensitivity; glucose tolerance or lipid levels) or included a chronic cardiometabolic condition (i.e. metabolic syndrome, cardiovascular disease or type 2 diabetes).

The exclusion criteria included the following: (i) the study included all three measures of interest but did not include them simultaneously in the statistical analyses; (ii) the study included only special populations (e.g. participants with type 1 diabetes); (iii) the study was not original research and/or (iv) the reviewers could not access the full text after contacting the corresponding author.

Identification of relevant studies

Two reviewers (EF, RL) independently reviewed all articles based on title and abstract initially, then assessed full text for eligibility. For articles that needed further clarification in order to assess their eligibility for the present review, the corresponding author was contacted. Any discrepancies between reviewers about article eligibility were discussed with all authors until a final consensus decision was reached.

Data extraction and coding

Data extraction was performed by one reviewer (EF) using a predetermined data extraction template. The following information was extracted from each article: (i) study design and length (for longitudinal studies only); (ii) participant characteristics (sample size, % males, age range, mean age, % overweight/obese and location); (iii) measures used to assess sedentary behaviour, dietary intake and cardiometabolic risk markers; (iv) statistical analysis methods (e.g. statistical models used, variables included in model) and (v) main findings in regard to sedentary behaviour, dietary intake and cardiometabolic risk markers. Where studies combined multiple domains of sedentary behaviours (e.g. television viewing, video watching and video playing), this was coded as 'screen time' in the results table. This method was also applied to dietary variables where multiple drinks were assessed and coded as 'sugar sweetened beverages'. For this review, only dietary vari-

ables related to food and drinks consumed, energy intake or macronutrient intakes were reported.

Methodological quality assessment

The methodological quality of the included studies was independently examined by two reviewers (EF, RL) using an adapted 15-item quality criteria checklist (26,27) (Table 1). The 15-item checklist consisted of assessing various methodological aspects (e.g. study design and sample, data sources and measurement of variables, statistical methods used) and involved a yes (0.5 or 1 point) or no/unclear (0 point) answer format. The change made to the original quality criteria checklist was mainly regarding the measurement questions, where the reliability and validity of the three measurements of interest were examined separately, as opposed to assessing the reliability and validity of one measurement. A quality score ranging from 0 to 15 points was calculated for each study. This score was

Table 1 Criteria list for assessment of the methodological quality of observational studies (based on Tooth *et al.* (26) and Singh *et al.* (27))

Criteria (rating of criteria: 0.5 or 1 = yes, 0 = no or unclear)	Score (total 15)
Study design	
1. Is the study design presented AND is the study design longitudinal and not cross-sectional?	__ / 0.5
Target population	
2. Do the authors describe the target population they wanted to research?	__ / 1
Sample	
3. Was a random sample of the target population taken/described AND/OR was the response rate 60% or more?	__ / 0.5
4. Is participant selection described, or referred to?	__ / 1
5. Is participant recruitment described, or referred to?	__ / 1
6. Are the inclusion and/or exclusion criteria stated, or referred to?	__ / 1
7. Is the study sample described? (minimum description = size, gender, age, BMI)	__ / 1
8. Are the numbers of participants at each stage of the study reported? (authors should report at least numbers eligible, numbers recruited and numbers with complete data)	__ / 1
Variables	
9. Are the measures of sedentary behaviour, dietary intake and the health outcome sufficiently described in detail?	__ / 1
Data sources and collection	
10. Do authors describe the source of their data? (e.g. registry, health survey) AND did authors describe how the data were collected? (e.g. by mail, by survey)	__ / 0.5
Measurements	
11. Was the validity of sedentary behaviour mentioned or referred to? AND/OR was the reliability of sedentary behaviour mentioned or referred to? (if measured by accelerometry, automatically full points)	__ / 0.5
12. Was the validity of dietary behaviour mentioned or referred to? (if measured by 24-h food recall, automatically full points)	__ / 0.5
13. Was the health outcome measured objectively and not by self-report?	__ / 1
Statistical methods	
14. Were appropriate statistical methods used and adequately described (including taking into account number of participants and clustering effects)? AND/OR did the statistical methods address confounders?	__ / 0.5
15. Were the numbers/percentages of participants with missing data for sitting and the health outcome indicated? AND if more than 20% of data in the primary analyses were missing, were methods used to address missing data?	__ / 0.5

divided by 15 points to calculate a percentage from 0 to 100% and a study was considered to be of high quality if the score was >50% (27).

Synthesis of studies

All findings relating to sedentary behaviour and cardiometabolic health from each study were categorized and tabulated into two categories: (i) no association between sedentary behaviour and the cardiometabolic risk marker (denoted by '0') or (ii) a significant positive or negative association between sedentary behaviour and cardiometabolic risk marker, independent of dietary intake (denoted by '+' or '-'). Following the 'quality rating system' by Singh *et al.* (27), the overall findings were synthesized into four levels of scientific evidence. However, because of all studies in this review being considered to be of 'high quality', cut-off percentages were given to differentiate the level of 'strong' and 'moderate' evidence. Thus, the four levels of scientific evidence used in this review are as follows: (i) *strong evidence* where ≥ 2 high-quality studies have $\geq 70\%$ consistent findings; (ii) *moderate evidence*

where ≥ 2 high-quality studies have between 50 and 69% of consistent findings; (iii) *inconsistent evidence* where ≥ 2 studies have <49% of consistent findings and (iv) *insufficient evidence* where only one study was available.

Results

Search results

The literature search identified a total of 539 records (Figure 1). After removing 12 duplicates and checking the titles, abstracts and full-text articles, 18 articles met the inclusion criteria. A further seven articles were identified from other sources (e.g. reference lists and personal libraries), resulting in a total of 25 articles being included in this systematic review.

Study characteristics

All 25 articles reported on 25 unique study samples. Twenty-one articles reported cross-sectional results and four reported longitudinal study results (ranging from 1 to

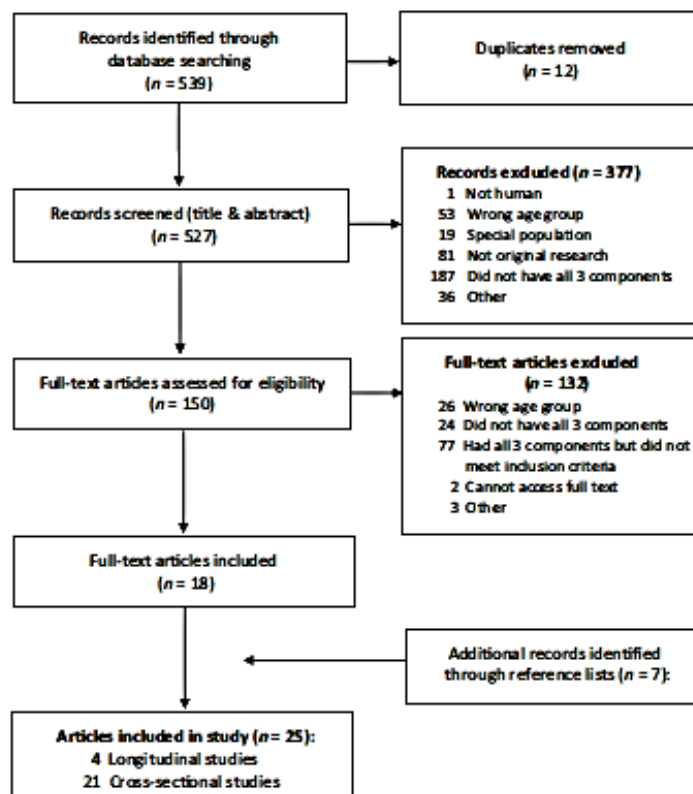


Figure 1 Flow diagram of the identification of literature for inclusion in this systematic review.

4 years of follow-up). The majority of studies originated from the United States ($n=9$), Canada ($n=3$) or China ($n=3$). The number of participants ranged from 282 to 39,011 with most studies including an approximately even ratio of males to females, and only one study including females only (28). The age of participants ranged from 10 to 19 years (mean 14.4 years) and the majority were of healthy weight, with the body mass index (BMI) ranging from 18 to 24 kg m⁻². Nearly all studies used some form of regression analyses (e.g. linear, logistic, longitudinal, multilevel) to examine the associations between sedentary behaviour and a cardiometabolic health outcome, adjusting for dietary intake in the model, with one study conducting a mediation analysis with dietary intake as the mediator (29). The characteristics and the main findings of studies are presented in the Supporting Information (Table S1).

Overview of measures

The most common sedentary behaviour measure was television viewing (29–41), followed by total screen time (28,30,42–49), personal computer use (31,32,36,37, 40,41), and video game playing or video viewing (37,40,49). Three studies measured total sedentary behaviour (28,50,51) and only one measured a non-screen time behaviour such as homework time (results not reported in the review) (31). The majority of sedentary behaviours were measured via a self-reported questionnaire, with two studies using a proxy report for those aged 12 or below (45,52), and one study each used a 24-h activity recall (28) or interview-administered questionnaire (47). The dietary variables that were included in the analyses varied widely, with the majority of studies ($n=19$) simultaneously adjusting for multiple dietary variables. The most common variables included total energy intake (34,35,40,41,46,47, 49–51) and the consumption of energy-dense snacks (29,30,32,36,39,42–45,48), sugar-sweetened beverages (28,30–33,36,37,44,45,50), fruits and vegetables (28,31,32,36,38,42,43,50), and fast food (30,42,45,50), with few studies measuring macronutrient intakes (28,35,40,47) or specific food items (e.g. cereals, meat) (50,52). Adiposity was the most commonly reported cardiometabolic risk marker, with 21 studies measuring adiposity mostly by weight status. Only four studies assessed other cardiometabolic markers such as blood pressure, insulin resistance, glucose tolerance and lipid levels (40,46–48).

Methodological quality assessment

A summary of the methodological quality of the studies is presented in the Supporting Information (Table S2). The initial discrepancy between the two reviewers when scoring

the 25 articles was 6.3%. The majority of disagreements were resolved after discussion, with 9 of the 33 discrepancies being discussed with all authors. The quality scores of the papers ranged from 60 to 84%. The most common reasons why studies did not get a quality score of 100% were the study 'did not use methods to address missing data' and 'did not report the inclusion and exclusion criteria of the sample'. According to the quality criteria (27), all studies were considered to be of high quality (>50%) with eight studies scoring 80% or more (28,30,31,41,42, 46,47,49).

Fifteen of the 25 studies included a random sample of the target population (29–34,36,38,39,41–43,45–48,50) with 14 studies reporting a response rate of 60% or more (29–31,33,34,38,41–44,46,47,49,50). All studies used either self-administered questionnaires or interviews to measure sedentary behaviour with four studies reporting the validity (28,29,48,49) and two studies reporting the reliability (28,48) of the measure used. In total, seven studies used a 24-h food recall to assess dietary intake (33–35,41,46,47,50), with five studies reporting the recall data from one occasion, and one study each reporting the recall data from either two (41) or three (35) occasions. Other commonly used measures were self-administered questionnaires ($n=10$) or food frequency questionnaires ($n=8$), with four studies reporting both the reliability and validity of these measures (31,40,44,49) and one study reporting only the validity (52). Out of the 21 studies that measured adiposity, nearly all studies used objective measures with only four studies using self-reported height and weight to calculate BMI (36,38,43,49). Four studies measured blood biochemistry profiles (40,46–48), with one of those studies reporting the risk of the metabolic syndrome (46). Regarding the analyses, only 10 studies both adequately described and used appropriate statistical methods (including taking into account clustering effects due to sampling design) and addressed missing data where relevant (28–31,36,41,42, 47–49).

Main findings

The associations between sedentary behaviour and cardiometabolic markers adjusting for dietary intake are reported in Tables 2 and 3. Because of the majority of studies reporting on adiposity outcomes, results for adiposity and other cardiometabolic outcomes were presented separately. Table 2 summarizes the associations between various screen time behaviours and total sedentary behaviour with measures of adiposity (e.g. BMI, waist circumference), adjusting for dietary intake. Table 3 summarizes the studies reporting on associations between screen time behaviours with the other cardiometabolic outcomes (e.g. blood pressure, blood glucose, lipids), adjusting for dietary intake.

Table 2 Associations between various sedentary behaviours and adiposity, adjusting for dietary intake

Author [quality %]	Dietary factors included in analysis*	TV	Video use or VG	PC	Total ST	Total SB
Arango (30) [80]	Soft drink, high-fat foods, fast foods	0 TV ^g WC +TV ^g WC			0 ST ^g WC + ST ^g BMI + ST ^g WC + ST ^g SR-BMI	
Berkey (49) [†] [80]	EI		+ Vid ^g SR-BMI			
Hsu (42) [80]	FV, sweets, snacks, fast food				+ ST ^g BMI	
Peart (41) [80]	EI, fat intake	+ TV ^g BMI		0 PC ^g BMI		
Tang (31) [80]	Soft drink, FV	+ TV ^g BMI		0 PC ^g BMI		
Must (28) [†] [80]	% CHO, protein and fat, FV, soft drink				0 ST ^g BMI 0 ST ^g WHR	0 SB ^g BMI
Nasreddine (50) [74]	EI, cereals, dairies, meat, legumes/nuts, FV, added fats/oils, fast food, sugar/sweets, SSBs					+ SB ^g BMI + SB ^g WC + SB ^g WHR
Al-Haifi (32) [74]	FV, SSBs (including energy drinks), milk, fast food, cakes/doughnuts, sweets	0 TV ^g BMI 0 TV ^g WC		0 PC ^g BMI 0 PC ^g WC + PC ^g BMI		
Kunesova (52) [74]	Fish intake/meat fat intake					
Forshee (33) [74]	All beverages (including SSBs, coffee/tea, beer)	0 TV ^g BMI				
Crespo (34) [74]	EI	+ TV ^g BMI				
Carson (29) [70]	Junk food (including sweets, soft drinks, cakes/pastries, potato chips, French fries)	+ TV ^g BMI				
Vik (43) [70]	FV, unhealthy snacks (including soft drinks, candy and potato chips)				+ ST ^g SR-BMI	
Zhang (35) [67]	EI, % fat intake	0 TV ^g BMI				
Fulton (51) [†] [64]	EI					0 SB ^g BMI + SB ^g BMI + SB ^g BMI
Hume (44) [64]	SSBs, unhealthy snacks (including soda, candy, potato chips)				0 ST ^g BMI 0 ST ^g SF 0 ST ^g WC + ST ^g WC	
Janssen (36) [64]	FV, sweets, soft drink, cake, potato chips	+ TV ^g SR-BMI		0 PC ^g SR-BMI		
Giammetti (37) [64]	Soft drink	+ TV ^g BMI	0 VG ^g BMI	0 PC ^g BMI		
Shan (45) [60]	Fast food, snacks, SSBs, alcohol				+ ST ^g BMI	
Delva (38) [60]	FV	+TV ^g SR-BMI				
Elger (39) [†] [60]	Snacks	+ TV ^g BMI				
Summary of results						
	0 No significant association	5	1	6	6	2
	+ Significant association, independent of diet	9	1	1	7	5

[†]Longitudinal study.

*Dietary variables adjusted for in the analyses involving the sedentary behaviour and health outcome variables.

%BF, % body fat; +, significant positive association, independent of diet; 0, no significant association; ^g, boys only; BMI, objectively measured BMI; EI, energy intake; FFM, fat-free mass; FMI, fat mass index; FV, fruits and vegetables; ^g, girls only; SF, skin-folds; SR-BMI, self-reported BMI; SSB, sugar-sweetened beverage; WC, waist circumference; WHR, waist to hip ratio.

As shown in Table 2, strong evidence was observed between computer use and adiposity. Six of the seven studies (86%) examining computer use found no significant association with adiposity, with the regression analyses adjusting mainly for soft drink, energy-dense snacks, and/or fruit and vegetable intake (31,32,36,37,41). Moderate evidence was found between both television viewing and total screen time with adiposity, with 64% (29–31,34,36–39,41) and 54% of the analyses (30,42–45,49) respectively finding a significant

and positive relationship, independent of dietary intake. However, no consistent pattern was observed according to the types of dietary variables adjusted for in the analyses. Additionally, there was moderate evidence to support a significant and positive relationship between total self-reported sedentary behaviour and adiposity (50,51), independent of total energy intake and other various food items. There was insufficient evidence of the relationship between video viewing and video game use (37,49).

Table 3 Associations between screen time behaviours with cardiometabolic risk markers and the metabolic syndrome, adjusting for dietary intake

Author	Dietary factors included in analysis*	Cardiometabolic risk markers							Met syndrome
		SBP	DBP	Insulin	Glucose	Total-C	LDL-C	HDL-C	
Kang (46) [84]	EI								0 [†] ST + [‡] ST
Sugiyama (47) [80]	EI, CHO, protein and fat intake	+ ST	0 ST						
Goldfield (40) [77]	EI, % fat intake	0 TV + VG 0 PC 0 ST	0 TV 0 VG 0 PC 0 ST			0 TV 0 VG 0 PC 0 ST	0 TV 0 VG 0 PC 0 ST	0 TV 0 PC 0 ST	0 TV + VG 0 PC 0 ST
Hardy (48) [77]	EDNP foods (including confectionery, hot chips, salty snacks)	0 ST [‡]		0 ST [‡]	0 ST [‡]		0 ST [‡]	0 ST [‡]	
Summary of results									
0 No significant association		4	5	1	1	4	5	4	3
+ Significant association, independent of diet		2	0	1	0	0	0	0	1

*Dietary variables adjusted for in the analyses involving the sedentary behaviour and health outcome variables.

+ , significant positive association, independent of diet; 0, no significant association; †, weekday only; ‡, weekend day only; ‡, boys only; CHO, carbohydrate intake; C-Ratio, cholesterol ratio (total cholesterol/HDL-cholesterol); DBP, diastolic blood pressure; EDNP, energy-dense nutritionally poor (foods); EI, energy intake; †, girls only; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; Total-C, total cholesterol.

When examining the findings from the four longitudinal studies, no consistent relationship was observed (28,39,49,51). Berkey *et al.* (49) and Elgar *et al.* (39) both reported positive longitudinal associations between screen time behaviours with adiposity, independent of total energy intake and intake of snacks, respectively; whereas Fulton *et al.* (51) and Must *et al.* (28) found no associations between overall sedentary behaviour and screen time with adiposity measures, independent of energy intake and percentage of macronutrient intake, respectively.

As shown in Table 3, three studies examined screen time behaviours with other cardiometabolic risk markers, such as blood pressure, insulin resistance, glucose tolerance and lipid levels (40,47,48), and one study examined total screen time and the risk of developing the metabolic syndrome (46). Overall, there was moderate (67%) to strong evidence (100%) to show no significant association between various screen time behaviours (e.g. television viewing, computer use) with systolic blood pressure (40,48), diastolic blood pressure (40,47), low-density lipoprotein (LDL)-cholesterol and high-density lipoprotein (HDL)-cholesterol (40,48). Because of the limited number of studies performed, there was insufficient evidence to draw conclusions on the remaining cardiometabolic risk markers as well as the association between screen time and the risk of metabolic syndrome (46).

Additionally, as there were a limited number of studies reporting the unadjusted and adjusted findings, it was not possible to draw conclusions on whether dietary intake attenuated the sedentary behaviour and health relationship. However, of the three studies that did report the unadjusted findings, screen time was no longer associated with dias-

tolic blood pressure (48) or the metabolic syndrome (46), and video game use was no longer negatively associated with HDL-cholesterol (40) after adjusting for either energy-dense, nutrient-poor foods (48) or total energy and macronutrient intake (40,46).

Discussion

This systematic review identified and summarized all studies published prior to March 2014 that have accounted for dietary intake when exploring the association between sedentary behaviour and cardiometabolic health markers in adolescents. According to the 15-item quality criteria checklist (26,27), all 25 studies identified were considered to be of high quality. From the 21 studies examining adiposity outcomes, there was moderate to strong evidence to show significant and positive associations between television viewing, screen time and overall sedentary behaviour with adiposity, independent of dietary intake. This is concerning since the majority of Western adolescents are already exceeding the screen time recommendations (5,11,12), thus placing them at an increased risk of becoming overweight or obese. However, findings from the four papers examining other cardiometabolic markers found no significant associations between screen time behaviours with blood pressure, LDL-cholesterol and HDL-cholesterol.

Most studies found television viewing was associated with obesity in adolescents, irrespective of dietary intake. These findings are similar to other reviews involving children and adolescents, where positive associations between television viewing and adiposity have consistently been

reported (6,53–56). For example, in the review by Tremblay *et al.*, among the 170 studies examining body composition, the majority of studies found positive associations between sedentary behaviour (mainly assessed by television viewing) and BMI (6). Similarly, findings from a meta-analysis revealed a small but significant relationship of 0.066 (95% CI= 0.056–0.078) between television viewing and body fatness among nearly 45,000 youth (53). However, although the majority of papers included in these reviews adjusted for physical activity, it is unknown whether the findings are also independent of dietary intake (6,53).

Although television viewing was shown to be independently associated with adiposity, computer use was not. This is consistent with previous literature involving children and adolescents, where they report a lack of association between computer use and obesity (53,54,57). There may be a couple of reasons for this difference. Firstly, television viewing is at the lowest end of the energy expenditure spectrum and requires less energy expenditure than computer use (58). Secondly, there is increased exposure to junk food advertising while watching television, as opposed to using a computer. This exposure to junk food advertising has been consistently linked with between-meal snacking (59). Further, 'mindless eating' where children pay less attention to how much they consume is more likely to occur while watching television as opposed to using the computer (60).

Of the four studies that examined those cardiometabolic markers other than adiposity, no associations were reported between screen time behaviours with blood pressure, LDL-cholesterol and HDL-cholesterol. This finding is in contrast to a previous review by Tremblay *et al.* who reported consistent evidence on the positive association between screen time and blood pressure and total cholesterol in school-age children and youth (6). However, it was not clear whether the studies included in that review adjusted for dietary covariates. A review by Chinapaw *et al.* reported similar findings to the current review where there was little evidence to draw conclusions on associations between 'sedentary time' (mainly measured by television viewing) and blood pressure or blood lipids (61).

The lack of findings in the present review could be attributed to a number of factors. It could be simply due to the limited number of studies available that have adjusted for dietary intake when reporting on cardiometabolic markers in young people. Alternatively, it could be due to the lack of lifetime exposure this age group has had to establish the risk markers for chronic disease, or to the lack of sensitive measures used in the studies (such as a one-off fasted blood test) to capture the small adverse effects in their cardiometabolic profile. Thus, for future studies examining cardiometabolic markers in children or adolescents, it is recommended to use more sensitive measures such as a

continuous glucose monitoring system or flow mediation dilation that are likely to detect the smaller fluctuations seen in a younger population group (62).

This review found that the majority of evidence relating to adiposity was independent of dietary intake. A possible explanation for this could be that the dietary variables adjusted for may not have been sufficient to attenuate the relationship. However, in the present review, because of the number of dietary variables examined across the included studies, it made it difficult to determine if independent (or null) associations were more commonly identified for one group of dietary variables over another (e.g. total energy intakes vs. macronutrient content). Additionally, because of the limited number of studies reporting the unadjusted and adjusted results (for dietary intake), it was not possible to draw conclusion as to whether a particular dietary variable attenuated the relationship (40,46,48). Further, it is also unknown whether these dietary variables reported were examined while participants were engaged in the screen time behaviours. If the behaviours were not measured concurrently, this suggests that dietary intake may be an indicator of an unhealthy lifestyle, rather than due to a direct association. Currently, there is limited research examining what dietary variables should be considered as covariates and what dietary variables, if any, mediate the relationship between sedentary behaviour and health in youth.

Another reason for the independent associations observed could be that dietary intake is simply not a strong driver of the relationship between screen time and adiposity. However, this explanation is at odds with previous literature where it is consistently reported that there are links between television viewing and elements of an unhealthy diet, such as lower fruit and vegetable consumption, higher consumption of energy-dense snacks, drinks, and fast foods, and higher total energy intake (20). However, there are mixed findings on whether the same unhealthy dietary behaviours are linked with other sedentary behaviours such as video game use (63–65) and objectively measured total sedentary time (66).

Limitations of the studies in the review

Despite the high quality of studies included in this review, it is important to acknowledge that there were several methodological limitations. Firstly, the majority of papers included in the review were cross-sectional, with the four longitudinal studies identified reporting inconsistent findings (28,39,49,51). Therefore, it is unknown whether sedentary behaviour and dietary intake are predictive of health outcomes longitudinally. Secondly, there were considerable variations in how sedentary behaviour and dietary intake were examined and assessed. All of the studies included in our review used subjective measures to assess sedentary

behaviour. Although these measures are useful to assess specific sedentary behaviour domains that could be targeted for interventions (e.g. reducing television viewing, or total screen time), they do not accurately capture the total time spent being sedentary. Thus, future studies are needed that also include an objective measure of overall sedentary time, such as accelerometers or inclinometers, to assess whether overall sedentary time has the same health implications as screen time behaviours.

Regarding dietary intake, there were two key limitations of the studies; the majority of studies only reported adjusted results and the studies reported on a vast array of dietary variables. By examining only the adjusted results, it is unknown whether the dietary variables attenuated the relationship between sedentary behaviour and adiposity. For future studies, it would be useful to report both the unadjusted and adjusted results when examining similar outcomes. Conducting mediation analyses would also help better understand the role of dietary intake in the sedentary behaviour and health relationship. For instance, was the relationship between sedentary behaviour and health explained partially or fully by dietary intake? The vast array of dietary variables made it difficult to identify common dietary variables adjusted for in the analyses among studies reporting positive vs. null findings. Future studies are needed examining the different types of dietary variables that may influence the sedentary behaviour and cardiometabolic health relationship. A suggested starting point would be to examine the dietary factors that have previously been shown to be associated with sedentary behaviour (e.g. total energy intake, fast food intake and snacking) (20) and adverse cardiometabolic health (e.g. energy-dense foods and sugar-sweetened beverages) (67–69). By examining specific dietary factors, it will provide much needed information on what dietary confounders to adjust for and thus provide greater consistency in future analyses.

Finally, there were inconsistencies in the analyses. For example, not all studies accounted for missing data where relevant or for clustering in their sampling design, thus potentially overestimating the statistical power of their findings. Additionally, not all studies adjusted for the same covariates, with only half of the studies adjusting for physical activity. However, it is interesting to note that there were no differences in findings between the studies that adjusted for physical activity and those studies that did not.

Strengths and limitations of review

Strengths of the review include broad search criteria examining many electronic sources, and a large number of studies were screened for eligibility by two reviewers. Additionally, an adapted quality assessment (26,27) was applied to distinguish low- and high-quality studies and was per-

formed independently by two reviewers. However, we acknowledge that the cut-off value to distinguish low and high quality is arbitrary. Other quality assessment tools such as the Effective Public Health Practice Project (70) and Downs & Black (71) were considered; however, these quality tools were mostly developed to assess randomized controlled trials and not observational studies. It is also possible that studies with significant findings were more likely to be included in the review. This was due to the inclusion criteria we applied to the review where studies were only included if they examined all three measures of interest in the analyses (sedentary behaviour, dietary intake and a cardiometabolic health outcome). However, if sedentary behaviour or dietary intake was previously found to be not significant in the initial correlation analyses with the health outcome, then typically these variables were not included in the regression analyses.

Conclusion

This systematic review found moderate to strong evidence of the relationships between self-reported television viewing, total screen time and overall sedentary behaviour with adiposity, independent of dietary intake. It is important to understand the nature of these independent relationships to help with informing the design of future interventions. For example, how much focus needs to be on reducing sedentary behaviours or whether targeting both sedentary behaviour and dietary intake would be more beneficial. However, further research is still needed to examine whether the independent associations remain when using objective measures to assess overall sedentary time. Additionally, exploratory and longitudinal studies would help with understanding whether these behaviours are occurring simultaneously, and whether one behaviour potentially influences the other over time.

Conflict of interest statement

The authors have no conflicts of interest to disclose.

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Supporting information

Additional Supporting Information may be found in the online version of this article, <http://dx.doi.org/10.1111/obr.12302>

Table S1. Description of the characteristics and main findings.

Table S2. Overall scores of the methodological quality assessment (ranked from highest to lowest).

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2.3 Manuscript supplementary files

The supplementary files supporting the manuscript “*Is the relationship between sedentary behaviour and cardiometabolic health in adolescents independent of dietary intake? A systematic review*” can be found on the following pages.

The supplementary files for this manuscript include:

- Table S1: Description of the characteristics and main findings of studies included in the review according to methodological quality
- Table S2: Overall scores of the methodological quality assessment (ranked from highest to lowest)

Table S1 - Description of the characteristics and main findings of studies included in the review according to methodological quality

Author, year; QS	Study population and follow-up	Sedentary behaviour measure	Dietary intake measure	Health outcome	Analyses conducted; adjustments	Results
Kang, 2010 ⁴⁶ ; 84%	n = 845, 53% boys, 10-18 yrs (mean 13.4) BMI 20.6, Korea	Total ST (incl TV, PC, internet) by wkday, wkend and total week (self-report)	Total EI (1x24HR)	MetS (BMI, WC, fasting glucose, lipids (objective))	Multivariate logistic regression (OR 95%CI), adjusted for age, sex, income, residence area, total EI and PA	+ assn: wkend ST (≥ 7 h) and MetS (OR 2.88 95%CI 1.04-7.98), adjusted for demographic variables, total EI and PA. No assn: wkday ST and overall week ST with MetS.
Arango ³⁰ , 2013; 80%	n = 546, 51% boys, 11-18 yrs (mean 14.4) BMI 19.2, Colombia	TV, PC, VG use (< or ≥ 2 h/d); Total ST (< or ≥ 3 h/d), by gender (self-report)	Daily intake soft drink, high fat foods, fast foods (questionnaire)	BMI z-s, WC, fitness (objective)	Linear regression model (B (SE)), adjusted for age, school, location, PA and diet variables	+ assn: >2h/d TV with WC (B=+0.22; p<0.02) (boys only) + assn >3h/d screen time with WC and BMI z-s (B=+0.34; p<0.01) (boys only), adjusted for diet
Berkey, 2003 ⁴⁹ ; 80%	n = 11,887, 43% boys, 10-15 yrs (mean NR) 19% overweight, USA	ST (incl TV, video and VG) and video only by gender (h/wk) (self-report)	Total EI (FFQ)	BMI (self-report)	Linear regression model (β , 95%CI); adjusted for EI, age, race, menarche history, tanner stage, height, and baseline BMI	Girls: + assn ST (TV, video and VG) (h/d) with change in 1-y BMI (+.045, CI .018-0.73) Boys: + assn video only with change in 1-y BMI (+0.07; CI .004-0.14)
Hsu, 2011 ⁴² ; 80%	n = 9,023, 47% boys, 7-11th grade (mean 15 yrs) BMI 20.8, China	ST (incl TV/video, PC) (min/d) (self-report)	Freq intake FV, sweets (eg desserts, ice-cream, candy, soda), snacks (eg potato chips), fast food (times/wk) (questionnaire)	BMI (objective)	Multivariate multilevel logistic modelling, controlling for VPA, SB, sleep, diet variables, health, parental edu/income, gender, school level	+ assn SB (min/d) and BMI (OR 1.11 95%CI 1.04-1.17), adjusting for all variables in model
Peart, 2011 ⁴¹ ; 80%	n = 2,368, 51% boys, 12-19 yrs (mean 15.4) 35% overweight/obese, USA	TV/video and PC use (≥ 2 h/d) (self-report)	Total EI and fat intake (2x24HR)	BMI (objective)	Multinomial logistic regression models (OR, 95%CI), adjusted for covariates in model (MPVA, diet) and gender, age, income and race	+ assn: TV/video (>2h/day) with overweight OR1.57 (CI 1.1-2.63) and obese OR1.84 (CI 1.24-2.69). No assn: PC (>2h/d) with overweight and obesity

Author, year; Q5	Study population and follow-up	Sedentary behaviour measure	Dietary intake measure	Health outcome	Analyses conducted; adjustments	Results
Tang, 2010 ³¹ ; 80%	n = 2,660, 50% boys, 11-16 yrs (mean NR) 14% overweight/obese, Vietnam	TV, PC/VG, study (≥ 2 h/wkday) (self-report)	Total EI, fast food, snacks, soft drinks, BF and FV intake (FFQ)	BMI (objective)	Hierarchical multivariate analyses (AOR, 95%CI), adjusted for residence location, household info, parental characteristics, and soft drink, FV, BF intake	+ assn: 2-3h (AOR1.9, CI 1.5-2.5) and >4h (AOR3.9, CI 1.5-9.9) TV (h/wkday) with BMI, adjusted for variables listed + assn: study time (after class) with BMI (AOR 1.5, CI 1.1-2.2) No assn: PC/VG with overweight
Must, 2007 ²⁸ ; 80%	n = 173, girls only 8-12 yrs base; follow-up 4+ yrs post-menarche BMI z-s 0.28, USA	Total SB (incl sleeping, lying, sitting and standing) (h/d); ST (h/d) (self-report)	% CHO, protein and fat, FV and soft drink intake (FFQ)	BMI z-s (objective) % BF (bio-impedance)	Linear mixed effects modelling, adjusted for age, parent overweight, and FV, soda and % protein intake	+ assn: total SB with %BF (+0.098, p=0.027), but not after adjusting for diet. No assn: ST with BMI z-s and %BF, or inactivity with BMI z-s
Sugiyama, 2007 ⁴⁷ ; 80%	n = 4,508, 51% boys, 12-19 yrs (mean 15.9) BMI 23.1, USA	Total ST (incl TV, video, PC and PC games (h/d) (self-report)	Total EI, macro- and micronutrient intake (1x24HR)	BMI, SBP, DBP (objective)	Multiple linear regressions (OR, 95%CI), adjusted for CHO and protein intake, BMI-z, race and income	+ assn: ST (h/d) with SBP (OR= .022; CI .007-0.38, p=.006) No assn: ST (h/d) with DBP
Goldfield, 2011 ⁴⁰ ; 77%	n = 282, 31% boys, 14-18 yrs (mean 15.5) BMI 34.7, Canada	TV, VG, PC (h/d) and calculated for total ST (h/d) (self-report)	Total EI, % fat intake (3-day FR)	BMI, BP (objective), lipids (fasting)	Multiple linear regression (B, 95% CI), adjusted for demographics, BMI, EI, % dietary fat, PA	+ assn: VG (h/d) and SBP (r=0.20, trig (r=0.10), C/HDL-C ratio (r=0.19) and neg assn HDL-C (r=0.19) (not adjusted). After adjusting variables: + assn VG and SBP (r=0.13, B=1.1) and total C/HDL ratio (r=0.12, B=0.10)
Hardy, 2010 ⁴⁸ ; 77%	n = 496, 58% boys, 15-16 yrs (mean 15.4) BMI 24, Australia	ST (incl TV, DVD, video, PC) (< or >2h/d) (self-report)	Usual intake soft drinks (< or >250ml/d), EDNP foods (daily/not daily) (FFQ)	BMI, WC, BP (obj), insulin, glucose, lipids (fasting)	Multiple logistic regression model (OR, 95%CI), adjusted for adiposity, SES, fitness, pubertal status, EDNP foods	+ assn >2h/d wkday ST and insulin (OR3.04) (p<0.05), after adjusting for WC, SES, EDNP foods, tanner score and CRE (boys)
Nasreddine, 2014 ⁵⁰ ; 74%	n = 498 aged 12-19 yrs*, 49.7% boys (total n in study 868), BMI 22.3, Lebanon *results reported for 12-19 participants only	Total SB (h/d) (self-report)	Total EI, cereals, dairies, meat, legumes/nuts, FV, added fats/oils, fast food, sugar/sweets, SSBs (1x24HR)	BMI z-s, WC, WHtR (objective)	Multivariate logistic regression analysis (OR, 95%CI), adjusted for baseline socio-demographics, BF intake, freq eating out, and PA	+ assn: SB (h/day) with overweight (OR 1.12, CI 1.03-1.21), obesity (OR 1.20, CI1.06-1.35), elevated WHtR (OR1.27, CI 1.13-1.43) and elevated WC (OR1.10, CI1.01-1.22)

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Author, year; QS	Study population and follow-up	Sedentary behaviour measure	Dietary intake measure	Health outcome	Analyses conducted; adjustments	Results
Al-Haifi, 2013 ³² ; 74%	n = 906, 51% boys, 14-19 yrs (mean 16.3), 49% overweight /obese, Kuwait	TV/video (TV), PC/internet (PC) (h/d) (self-report)	Weekly intake BF, SSBs (incl energy drinks), FV, milk, cakes/doughnuts, sweets, fast food (questionnaire)	BMI, WC (objective)	General linear model (partial eta squared), adjusted for PA, SB and diet	No assn: TV and PC (h/d) with BMI or WC
Kunesova, 2007 ⁵² ; 74%	N = 1,427 (aged 6-18 years) <small>Results reported separated for 13-18 years (n = 715)</small> NR% boys, (mean NR) NR, Czech Republic	TV, PC/games, reading, drawing (h/wk) (self-report)	Fish and meat fat intake (FFQ via interview)	BMI (objective)	Multiple linear regression, backward reduction of insignificant factors (B, SE), adjusting for gender, parent obesity, age, fish intake, fat meat intake, PA and house work	+ assn: time spent PC with BMI (B = 0.145, SE 0.061, p= 0.017), adjusting for variables listed
Forshee, 2004 ³³ ; 74%	n = 2,216, 52% boys, 12-16 yrs (mean 14) BMI 21.7, USA	TV on prev day (h/d as continuous variable) (self-report)	Bev intake: fruit drinks, diet/reg soft drinks, coffee, tea, and beer (FFQ per mth and 1x24HR)	BMI (objective)	Multivariate regression models (Coefficient, 95% CI), adjusted for beverage intake, PA and demographics	No assn: TV and BMI in both FFQ model and 24HR model
Crespo, 2001 ³⁴ ; 74%	n = 4,069, 49% boys, 8-16 yrs (mean 12) 12% obese, USA	TV use on prev day, categorised <1, 2, 3, 4 or >5h/day (self-report)	Total EI (1x24HR)	BMI (objective)	Proc Rlogist from Sudaan (RR, 95% CI), adjusted for age, income, EI, PA	+ assn: TV (2,3,4 and 5 h/day) with BMI, independent of total EI. Those watching >5/day most at risk (boys OR 2.63, 95%CI 1.01-6.85; girls OR 2.53, 95%CI 1.01-6.36)
Carson, 2012 ²⁹ ; 70%	n = 15,973, 48% boys, 12-16 yrs (mean 14.1) 24% overweight /obese, Canada	TV use (h/d), categorised into quartiles (self-report)	Freq of junk food intake (incl sweets, soft drinks, cake, potatoes, chips, french fries) (never to everyday) (questionnaire)	BMI z-s (objective)	Linear regression and multiple mediation analyses (B, 95% CI), adjusted for gender, age, race, family structure, SES, diet variables	+ assn: TV and BMI (Q4 B=0.22 CI 0.14-0.31), independent of TV snacking and junk food intake

Author, year; QS	Study population and follow-up	Sedentary behaviour measure	Dietary intake measure	Health outcome	Analyses conducted; adjustments	Results
Vik, 2010 ⁴³ ; 70%	n = 2,870, 51% boys, 15-16 yrs (mean 15.5) 24% overweight/obese, Norway	ST incl TV/PC use (h/d after school) (self-report)	FV and unhealthy snack intake (incl soft drinks, candy and potato chips) (questionnaire)	BMI (self-report)	Multivariate logistic regression analyse (OR, 95% CI), adjusted for meals eaten, gender, education plans, FV, unhealthy snacks and PA (model 4), and dieting (model 5)	+ assn: TV/PC (h/wk) with BMI (OR 1.09, CI 1.00-1.17), after adjusting for Model 4 variables. After adjusting for dieting (model 5), no longer significant
Zhang, 2010 ³⁵ ; 67%	n = 967 aged 12-18 yrs, 54% boys (total n study = 1,977), 6.2% overweight, China	TV and study (h/d) (self-report)	Total EI, fat intake (grams, %EI) (3x24HR)	BMI (objective)	Regression analyses (R ²), adjusted for survey year, gender, region, family income, diet, PA and TV	No assn: TV (h/d) and study (h/d) with change in BMI from 1997 to 2000 or 1997 to 2004, adjusting for variables listed
Fulton, 2009 ³¹ ; 64%	n = 472, 48% boys, 8-14 yrs (mean 10.6 base; 14.4 follow-up) BMI 20, USA	Total SB (incl TV, reading, PC) (100min/d) (self-report)	Total EI (FFQ)	BMI, FFMI, FMI (objective)	Longitudinal multilevel modelling, EI, SB, MVPA, gender and race	+ assn: for every 100min/d of SB, BMI and FFM increased .10kg/m ² , adjusted for EI, MVPA, gender and race No assn: SB and FMI
Hume, 2009 ⁴⁴ ; 64%	n = 580, 48% boys, 12-13 yrs (mean 12.75) BMI 18.6, Netherlands	ST (TV and PC), by gender (h/d) (self-report)	Ave daily intake of SSBs and high calorie snack foods (incl soda, candy, potato chips) (prev week) (questionnaire)	BMI, WC, skinfolds (objective)	Logistic regression, adjusted for ethnicity, organised sport, intake of SSBs and snacks, and clustering by school	+ assn: 3h/d SB (OR 3.4, 95%CI 1.1-10.7) and >4h/d SB (OR 5.5, 95%CI 2.1-14.1) with WC in girls only. No significant assn among boys
Janssen, 2004 ³⁶ ; 64%	n = 5,890, 48% boys, 11-16 yrs (mean NR) BMI 20.2, Canada	TV/video and PC use, by gender on typical wkday and wkend (h/d) (self-report)	Ave weekly intake of FV, sweets, soft drinks, cake/pastries, chips (questionnaire)	BMI (self-report)	Multiple logistic regression (OR 95%CI), adjusted for age, dietary variables and leisure time activities (e.g. PA, TV)	+ assn: TV (h/d) and BMI, with increasing TV time having greater odds of overweight and obese in both girls and boys (OR ranged 1.58 to 2.42) No assn PC (h/d) and BMI (p>0.1)
Giammette, 2003 ³⁷ ; 64%	n = 305, 48% boys, 11-14 yrs (mean 12.6) BMI z-s 0.57, USA	TV, PC, VG (>2h/d) (self-report)	Daily intake of reg/diet soft drinks (questionnaire)	BMI (objective)	Logistic regression (parameter estimate/OR, SE), adjusted for soft drink intake, age and sex	+ assn: >2h/d TV and BMI >85 th (OR 1.5 CI 1.17-1.93), after adjusting for soft drink, age and sex. After adjusting for ethnicity, no longer significant.

Author, year; QS	Study population and follow-up	Sedentary behaviour measure	Dietary intake measure	Health outcome	Analyses conducted; adjustments	Results
Shan, 2010 ⁴⁵ ; 60%	n = 19,517 aged 6-18 yrs, 50% boys (total n in study 21,198) 22% overweight/ obese, China	Total ST (≥2h/d) (proxy and self-report)	Alcohol, SSBs, snack food and fast food intake (times/wk) (questionnaire)	BMI (objective)	Multinomial logistic regression models (AOR, 95% CI), adjusted for covariates in model (smoking, diet variables, sleep) and age, gender, tanner stage and residence location	No assn: ST (h/d) and overweight + assn: ST 2-3h/d (AOR 1.27, CI 1.12-1.44) and ST >4h/day (AOR 1.32, CI 1.07-1.62) with obesity, adjusted for variables listed
Delva, 2007 ³⁸ ; 60%	n = 39,011, 46% boys, 13-16 yrs (mean 14.5) 25% overweight/ obese, USA	TV use wkday and wkend, by gender (h/d) (self-report)	Freq intake of green FV (never to everyday) (FFQ)	BMI (self-report)	Multiple logistic regression (OR, 95% CI), adjusted for race, population density, region, eating freq, sleeping freq, PA and parenting	+ assn: TV freq with BMI, adjusting for diet (boys OR 1.09 CI 1.11-1.18; girls OR 1.15 CI 1.11-1.18)
Elgar, 2005 ³⁹ ; 60%	n = 355, 45% boys, 11-17 yrs (mean 12.3 base; 15.4 follow-up) BMI 20, Wales	TV/video, PC games (h/wk) (self-report)	Freq snacks/day intake (questionnaire)	BMI (objective)	Multiple regression analyses (B, SE), adjusted for demographics, sports, meal skipping and snacks/day	+ assn: TV in yr 7 related to BMI in yr 11 (B=0.15, P=0.006), adjusting for demographic variables and snacks/day. No sig effect after adjusting for baseline BMI

QS: Quality score, NR: not reported, incl: includes, TV: television viewing, PC: computer use, VG: video game use, ST: total screen time, SB: sedentary behaviour, h/d: hours per day, wkday: weekday, wkend: weekend, BF: breakfast, FV: fruit and vegetable, SSBs: sugar sweetened beverages, EI: energy intake, bev: beverage, EDNP: energy-dense nutrient poor (foods), CHO: carbohydrates, FFQ: food frequency questionnaire, 24HR: 24 hour food recall, BMI: body mass index, z-s: z-score, WC: waist circumference, FFM: fat free mass, FFMI: fat free mass index, BF: body fat, BP: blood pressure, SBP: systolic blood pressure, DBP: diastolic blood pressure, PA: physical activity, MVPA: moderate to vigorous physical activity, SES: socio-economic status, OR: odds ratio, AOR: adjusted odds ratio, CI: confidence interval, SE: standard error, + assn: significant positive association, - assn: significant negative association, no assn: no significant association.

Table S2 - Overall scores of the methodological quality assessment (ranked from highest to lowest)

Author/criteria	Study design		Sample					Variables and Data source			Measurement			Statistical methods		Total (out of 15)	%	
	Target pop.		1	2	3	4	5	6	7	8	9	10	11	12	13			14
Kang, 2010 ⁴⁶	0	1	1	1	1	1	1	1	1	1	1	0	1	1	0.5	1	12.5	84
Arango, 2013 ³⁰	0	1	1	1	1	1	1	1	1	1	1	0	0	1	1	1	12	80
Berkey, 2003 ⁴⁹	1	0	0.5	1	1	1	1	1	1	1	1	0.5	1	0	1	1	12	80
Hsu, 2011 ⁴²	0	1	1	1	1	1	1	1	1	1	1	0	0	1	1	1	12	80
Peart, 2011 ⁴¹	0	1	1	1	1	1	1	1	0	1	1	0	1	1	1	1	12	80
Tang, 2010 ³¹	0	1	1	1	1	1	1	1	1	0	1	0	1	1	1	1	12	80
Must, 2007 ²⁸	1	1	0	1	1	1	1	1	0	1	1	1	0	1	1	1	12	80
Sugiyama, 2007 ⁴⁷	0	1	1	1	1	1	1	1	0	1	1	0	1	1	1	1	12	80
Goldfield, 2011 ⁴⁰	0	1	0	1	1	1	1	1	1	1	1	0	1	1	1	0.5	11.5	77
Hardy, 2010 ⁴⁸	0	1	0.5	1	1	1	1	1	0	1	1	1	0	1	1	1	11.5	77
Nasreddine, 2014 ⁵⁰	0	1	1	1	1	1	1	1	1	0	1	0	1	1	0	1	11	74
Al-Haifi, 2013 ³²	0	1	0.5	1	1	1	1	1	1	1	1	0	0	1	0.5	1	11	74
Kunesova, 2007 ⁵²	0	1	0	1	1	1	1	1	1	1	1	0	0.5	1	0.5	1	11	74
Forshee, 2004 ³³	0	1	1	1	1	1	1	1	0	1	1	0	1	1	1	0	11	74
Crespo, 2001 ³⁴	0	1	1	1	1	1	1	1	0	1	1	0	1	1	1	0	11	74
Carson, 2012 ²⁹	0	0	1	1	1	1	1	1	1	1	1	0.5	0	0	1	1	10.5	70
Vik, 2010 ⁴³	0	1	1	1	1	1	1	1	1	1	1	0	0	0	1	0.5	10.5	70
Zhang, 2010 ³⁵	0	1	0.5	1	1	1	1	1	0	0	1	0	1	1	0.5	1	10	67
Fulton, 2009 ⁵¹	1	1	0	0	1	1	1	1	0	1	1	0	0	1	1	0.5	9.5	64
Hume, 2009 ⁴⁴	0	0	0.5	1	1	1	1	1	0	1	1	0	1	1	1	0	9.5	64
Janssen, 2004 ³⁶	0	1	0.5	1	1	1	1	1	0	1	1	0	0	0	1	1	9.5	64
Giammettei, 2003 ³⁷	0	1	0	1	1	1	1	1	1	0	1	0	0	1	0.5	1	9.5	64
Shan, 2010 ⁴⁵	0	1	0.5	1	1	1	1	1	0	0	1	0	0	1	0.5	1	9	60
Delva, 2007 ³⁸	0	1	1	1	1	1	1	1	0	1	1	0	0	0	1	0	9	60
Elgar, 2005 ³⁹	1	1	0.5	1	1	1	1	1	0	0	1	0	0	1	0.5	0	9	60

2.4 Summary

This systematic review identified and summarised the current literature that has either performed mediating analyses or accounted for dietary intake when exploring the associations between sedentary behaviour and cardiometabolic health markers in adolescents. Overall, the majority of studies included in the review found significant positive associations between TV viewing, screen time and self-reported total sedentary behaviour with markers of adiposity, irrespective of dietary intake. The systematic review also highlighted a number of gaps in the literature, in particular, the limited number of studies available that have examined relationships between sedentary behaviour and cardiometabolic risk markers other than adiposity (e.g. blood pressure, glucose, inulin) and the lack of objective measures used to assess sedentary time (e.g. accelerometry). The review also highlighted the inconsistency of dietary variables examined in the studies. This could be evidence of convergent validity, thus making it difficult to assess the overall impact of specific dietary factors on the sedentary behaviour and cardiometabolic health relationship.

Based on the findings from this systematic review, the thesis has been structured accordingly to address the identified gaps in the literature. These include: examining the relationship between sedentary behaviour and cardiometabolic health in adolescents using a combination of self-reported and objective measures of sedentary behaviour; investigating key dietary variables that could potentially mediate the sedentary behaviour and cardiometabolic health relationship; and examining a number of cardiometabolic outcomes, including measures of adiposity (e.g. BMI), metabolic syndrome and individual cardiometabolic markers such as glucose. The

next Chapter will outline the overall aim of the thesis, the specific objectives and hypotheses for each study and highlight the significance of the potential findings of this thesis.

CHAPTER THREE: RESEARCH AIMS

CHAPTER 3: RESEARCH AIMS

3.1 Aims and objectives

The overall aim of this thesis is to explore the role of dietary intake in the relationship between sedentary behaviour and cardiometabolic risk markers in adolescents. Specifically, the thesis has four main objectives that address the gaps outlined previously:

- 1) Examine the cross-sectional mediating effects of dietary intake on the relationships between self-reported sedentary behaviour (e.g. TV viewing), BMI and metabolic syndrome in U.S. adolescents (*Study 1*)
- 2) Examine the cross-sectional mediating effects of dietary intake on the relationships between objectively-measured sedentary behaviour (e.g. total volume and bouts), BMI and metabolic syndrome in U.S. adolescents (*Study 2*)
- 3) Examine the cross-sectional and prospective mediating effects of dietary intake on the relationship between sedentary behaviour (both self-reported and objectively-measured) and BMI in Australian adolescents (*Study 3*)
- 4) In an experimental study, examine the role of dietary intake (high-energy diet *versus* standard-energy diet) on the relationship between uninterrupted sitting, sitting with resistance-type activity breaks and postprandial glucose in healthy Australian adolescents (*Study 4*)

3.2 Hypotheses

The overarching hypothesis of this thesis is that dietary intake partially mediates the relationship between TV viewing and the cardiometabolic health outcomes

examined, both cross-sectionally and prospectively. However, it is hypothesised that diet would not mediate the relationship when objectively assessed total volume or bouts of sedentary time are examined.

Study 1: Mediating effects of diet on associations of TV viewing, body mass index, and metabolic syndrome in adolescents

Study 1 examined whether five different elements of dietary intake (e.g. total energy intake, fruit and vegetable intake, discretionary snacking, sugar-sweetened beverage consumption, and dietary quality) mediate the relationships between TV viewing, BMI and metabolic syndrome in a large sample of U.S. adolescents. It was hypothesised that a lower intake of fruit and vegetables and a lower dietary quality score, and higher intakes of total energy, discretionary snacks and sugar-sweetened beverages would mediate the positive relationships found between TV viewing with BMI, and TV viewing with metabolic syndrome.

Study 2: Mediating effects of diet on associations of volume and bouts of sedentary time and cardiometabolic health in adolescents

Study 2 examined whether five different elements of dietary intake (e.g. total energy intake, discretionary snacking, sugar-sweetened beverage consumption, fruit and vegetable intake and dietary quality) mediated the associations between objectively assessed total volume and bouts of sedentary time with BMI and metabolic syndrome in U.S. adolescents. It was hypothesised that none of the dietary intake variables would mediate the positive relationships found between total volume and bouts of sedentary time with BMI and metabolic syndrome in U.S. adolescents.

Study 3: Cross-sectional and prospective mediating effects of diet on associations of sedentary behaviour and body mass index in adolescents

Study 3 examined the cross-sectional and prospective mediating effects of the frequency of consuming discretionary snacks, sugar-sweetened beverages and takeaway foods on the association of TV viewing with BMI in Australian adolescents. It was hypothesised that the consumption of discretionary snacks, sugar-sweetened beverages and takeaway foods would partially mediate the cross-sectional and prospective associations between TV viewing and BMI, but would not mediate the cross-sectional or prospective associations between total volume and bouts of sedentary time with BMI.

Study 4: Effects of breaking up sitting on adolescents' postprandial glucose after consuming meals varying in energy: an experimental trial

Study 4 examined the impact of uninterrupted sitting *versus* sitting with resistance-type activity breaks on adolescents' postprandial interstitial glucose responses while consuming a high-energy diet and standard-energy diet. It was hypothesised that the activity breaks would significantly attenuate postprandial glucose compared to uninterrupted sitting for both meal conditions, but would have a greater attenuation in the high-energy diet condition.

3.3 Significance

This thesis presents unique epidemiological data on the role that dietary intake plays in the associations between various sedentary behaviours and cardiometabolic risk markers. It also presents unique experimental data on the role that dietary intake plays on postprandial glucose when sitting is prolonged or when sitting is interrupted with postprandial glucose in a controlled setting. The findings from this thesis are

important for a number of reasons. Firstly, the findings may help to understand the current discrepancies in the adolescent literature. For example, as this thesis specifically examines the mediating role of dietary intake in various relationships between sedentary behaviour and cardiometabolic health, it will help to understand whether the consistent associations observed between TV viewing and BMI are driven partially by dietary intake. Further, this thesis explores other cardiometabolic risk markers (e.g. metabolic syndrome and postprandial glucose) which will add key health evidence in an area that is currently lacking.

Secondly, the findings may inform future interventions to establish how much focus needs to be on changing certain lifestyle behaviours. For example, if dietary intake is shown to be a mediator between TV viewing and obesity, targeting a reduction in TV viewing time may subsequently reduce energy intake and discretionary snacking. In contrast, if diet is not found to play a key role in the sedentary behaviour and health relationship, these findings will further highlight the adverse independent effects sedentary behaviour has on cardiometabolic health.

Thirdly, findings from this thesis may help contribute evidence to inform future public health guidelines. Currently, the sedentary behaviour guidelines for adolescents recommend breaking up long periods of sitting as often as possible (as well as recommending screen time being limited ≤ 2 hour/day).¹ However, to date, there are no guidelines on the ideal time to break up sitting (e.g. before or after consuming a main meal) or on how much and how long we should be breaking up sitting time. Findings from the experimental study (*Study 4*) may help to contribute novel evidence on whether there are additional health benefits in interrupting sitting time when performed after consuming a main meal.

Lastly, findings from this thesis contribute further evidence to the sedentary behaviour research area, which overall, may have a significant impact at the population level. For example, given sedentary behaviour² and dietary intake³ are often established at a young age, any health related behaviour changes that can be made during adolescence (whether it be small or large) may reduce the risk of developing lifestyle-related chronic health conditions during adulthood. Therefore, by preventing or tackling the issue during adolescents, this may have the potential for future public health benefits through reducing the burden of disease.

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CHAPTER FOUR: STUDY ONE

MEDIATING EFFECTS OF DIET ON
ASSOCIATIONS OF TV VIEWING, BODY MASS
INDEX, AND METABOLIC SYNDROME
IN ADOLESCENTS

CHAPTER FOUR: STUDY ONE

MEDIATING EFFECTS OF DIET ON ASSOCIATIONS OF TV VIEWING, BODY MASS INDEX, AND METABOLIC SYNDROME IN ADOLESCENTS

4.1 Overview

The systematic review in Chapter Two found only one study had specifically used mediation analyses to examine the role of dietary intake in the associations between TV viewing time and BMI, with the other studies adjusting for dietary intake in a regression analyses. The lack of studies performing mediation analyses makes it difficult to understand how much, if any, dietary intake is on the explanatory pathway between sedentary behaviour and cardiometabolic health. In addition, the majority of studies assessed markers of adiposity as the outcome, with very few studies examining other cardiometabolic markers. Therefore, the aim of this study was to explore the mediating effects of dietary intake on the relationship between TV viewing and two key cardiometabolic health outcomes: BMI and metabolic syndrome.

The following study has been published as a manuscript in *Obesity Science & Practice* (a new journal affiliated with the journals *Obesity*, *Pediatric Obesity and Obesity Reviews*) and has been formatted according to their requirements. The citation for the manuscript is: *Fletcher EA, McNaughton SA, Lacy KE, Dunstan DW, Carson V, Salmon, J. Mediating effects of dietary intake on associations of TV viewing, body mass index and metabolic syndrome in adolescents. Obesity Science & Practice. 2016;2: 232-240.* The manuscript supplementary files can be found directly

after the manuscript in *Section 4.3*. The ‘Authorship Statement’ for this manuscript and a copy of the ‘Permission Statement to Publish’ this manuscript are contained in Appendix C.1 and Appendix C.2, respectively.

Findings from this manuscript were presented at the International Society of Behavioral Nutrition and Physical Activity conference in Cape Town, South Africa in June 2016 and at the 11th Research Degree Symposium held by the School of Exercise and Nutrition Sciences, Deakin University in Melbourne, Australia in September 2015 (abstracts available in Appendix C.3 and Appendix C.4, respectively).

The author contributions to the manuscript were as follows, with the candidate contributing to all aspects of the manuscript process: Fletcher, McNaughton, Dunstan, Lacy and Salmon designed the study; Fletcher calculated the main variables used in the analyses with guidance from McNaughton in calculating the dietary data; Fletcher performed the main analyses with guidance from Carson; Fletcher wrote the initial draft of the manuscript; and McNaughton, Lacy, Dunstan, Carson and Salmon provided critical edits and editions to the manuscript. All authors approved the final manuscript.

4.2 Manuscript

ORIGINAL ARTICLE

Mediating effects of dietary intake on associations of TV viewing, body mass index and metabolic syndrome in adolescents

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Summary**Objective**

Evidence suggests that TV viewing is associated with body mass index (BMI) and metabolic syndrome (MetS) in adolescents. However, it is unclear whether dietary intake mediates these relationships.

Methods

A cross-sectional analysis was conducted in adolescents (12–19 years) participating in the 2003–2006 United States National Health and Nutrition Examination Survey. BMI z scores (zBMI) ($n = 3,161$) and MetS ($n = 1,379$) were calculated using age- and sex-specific criteria for adolescents. TV viewing (h/day) was measured via a self-reported questionnaire, and dietary intake was assessed using two 24-h recalls. Using the MacKinnon method, a series of mediation analyses were conducted examining five dietary mediators (total energy intake, fruit and vegetable intake, discretionary snacks, sugar-sweetened beverages and diet quality) of the relationships between TV viewing and zBMI and MetS.

Results

Small positive relationships were observed between TV viewing and zBMI ($\beta = 0.99$, $p < 0.001$) and TV viewing and MetS (OR = 1.18, $p = 0.046$). No dietary element appeared to mediate the relationship between TV viewing and zBMI. However, sugar-sweetened beverage consumption and fruit and vegetable intake partially mediated the relationship between TV viewing and MetS, explaining 8.7% and 4.1% of the relationship, respectively.

Conclusions

These findings highlight the complexity of the relationships between TV viewing, dietary intake and cardiometabolic health outcomes, and that TV viewing should remain a target for interventions.

Keywords: BMI, dietary intake, metabolic syndrome, television.

Introduction

Currently, 17% of adolescents from the United States (US) are obese, and 5.8% have extreme obesity (1). Concerningly, among obese adolescents, 30% have the metabolic syndrome (MetS) (2), a condition characterized by a collection of risk factors that increase a person's chances of developing type 2 diabetes and cardiovascular disease (3). Because obesity has been shown to track from adolescence to adulthood (4) and obesity-related

behaviours are often established at a young age (5,6), it is imperative to understand the key drivers of adolescent obesity and MetS to help inform effective interventions.

Time spent watching TV has consistently been shown to have adverse links with adolescent obesity (7) and MetS (8). However, the exact mechanism underlying these associations remains unclear. It has been suggested that TV viewing may displace time spent in physical activity. However, a meta-analysis of 163 studies in children and adolescents found that the association

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between TV viewing and physical activity was very small ($r = -0.108$), implying that children and adolescents may have both high amounts of TV viewing and physical activity (9). Alternatively, TV viewing has been consistently linked to elements of an unhealthy diet such as a higher energy intake, a greater intake of sugar-sweetened beverages (SSB) and snacks, lower intakes of fruit and vegetables, and a poorer overall diet (10–13). These unhealthy dietary elements are also important risk factors for obesity (14) and MetS (15).

Therefore, this poses the question as to whether the associations found between TV viewing, body mass index (BMI) and MetS could be partially explained by elements of a less healthy diet. If dietary intake is found to partially mediate these relationships, these findings will help to inform future interventions that targeting both TV viewing and dietary intake may have a greater impact on reducing obesity or MetS prevalence rather than targeting these behaviours individually. To date, only one study has specifically explored the mediating role of diet in an adolescent population and found TV snacking and junk food consumption did not mediate the relationship between TV viewing and BMI (16). However, this study was limited by only examining two dietary mediators, and only explored the relationship between TV viewing and BMI.

Therefore, the present study aims to identify whether five different elements of dietary intake (e.g. total energy intake, fruit and vegetable intake, discretionary snacking, SSB consumption and dietary quality) may partially mediate the relationships between TV viewing, BMI and MetS in a large sample of US adolescents. It is hypothesized that a lower intake of fruit and vegetables and dietary quality score, and a higher intake of total energy, discretionary snacks and SSB will mediate the relationships between TV viewing with BMI, and TV viewing with MetS.

Methods

The National Health and Nutrition Examination Survey conducted in 2003–2004 and 2005–2006 had a multi-stage, stratified sampling design used to recruit a representative cross-sectional sample of the US population. The 2003–2004 and 2005–2006 years were selected because of having the same dietary variables in both datasets and availability of the accelerometer data. Ethics was approved by the National Center for Health Statistics Institutional Review Board and Research Ethics Review Board (Protocol #98-12). Full details of the data-collection procedures are available elsewhere (17). In total, 5,138 adolescents aged 12–19 years were initially contacted and invited to participate in the study. Of those approached, 4,591 adolescents provided consent and completed a questionnaire on their demographics and

activity/sedentary behaviour levels. Afterwards, 4,455 adolescents attended a health examination visit to collect physiological measurements and to complete one of two 24-h dietary recall. The same participants were then followed up 4 to 11 days later to complete the second 24-h dietary recall.

Cardiometabolic measurements

Height, weight, waist circumference, blood pressure, fasting blood glucose and insulin, and serum lipids were measured from the participants who attended the health examination visit. The full protocol and assessment of anthropometric measures and blood analyses can be found elsewhere (18,19). Briefly, height and weight were measured in a light gown without shoes by a research nurse, and waist circumference was measured using a flexible steel measuring tape underneath clothing at the uppermost lateral border of the ilium crest. Height and weight were used to calculate each participant's BMI z scores (zBMI) using age- and sex-specific BMI percentiles (STATA, SJ13-2 dm0004_1) based on the 2000 Centers for Disease Control and Prevention US growth charts (20). Waist circumference, systolic and diastolic blood pressure, blood glucose and insulin, and serum lipids were used to calculate the presence of MetS following the age- and sex-specific criteria specifically designed for adolescents set by the International Diabetes Federation. According to these criteria an adolescent (aged 12–19 years) has MetS if he or she has an elevated waist circumference and two or more of the following four criteria: elevated systolic or diastolic blood pressure, low HDL-cholesterol, elevated triglycerides and elevated plasma glucose (3).

Television viewing

The amount of time spent watching TV was self-reported via the question 'Over the past 30 days, on average about how many hours did you sit and watch TV or videos?'. The response options were: none, less than 1 h, 1 h, 2 h, 3 h, 4 h and 5 h or more. Similar questions have been shown to be reliable in youth (21). For all analyses, TV viewing was treated as a continuous variable.

Dietary intake mediators

Dietary intake was assessed via two interviewer-administered 24-h dietary recalls, delivered via a computer-assisted system (the Automated Multiple-Pass Method) (22). Participants aged 12 years and older reported their first dietary recall at the health examination visit, and the second dietary recall was conducted 4 to 11 days later by telephone. All dietary data were coded

using the USDA National Nutrient Database for Standard Reference (23).

Based on previous associations with TV viewing time (11,12,24,25), the dietary intake elements examined as mediators included: (i) total energy intake; (ii) fruit and vegetable consumption; (iii) discretionary snack consumption; (iv) SSB consumption and (v) diet quality. All dietary intake elements were calculated based on the average of two days of dietary recall data. Total energy intake (calories) was calculated for each participant based on the quantity of food and beverages reported. Daily servings of fruit and vegetables were calculated using the Food Patterns Equivalents Database (26). Fruit was defined as any whole fruit (not including fruit juice) and vegetables included potatoes, beans and legumes. A serving of fruit or vegetables was equivalent to one cup of raw, canned or frozen fruit or vegetables, or two cups of leafy green vegetables (26).

Discretionary snacks were defined as grain-based and dairy-based desserts, cereal-, protein-, granola- and other-bars, sweet snacks and candies, sugar-syrups and preserves, salty snacks from grain or starchy vegetables and dips/spreads. Artificially sweetened snacks were excluded (27). SSB were defined as any non-alcoholic beverage with added sugar including soda, fruit-flavoured drinks (not 100% juice), sweetened tea, coffee and milk drinks, sport drinks and energy drinks (28). Any 'diet' drinks, 100% fruit juice, or unsweetened tea or coffee were not included as a SSB. A serving of discretionary snacks and a serving of SSB was equivalent to 143 calories (600 kilojoules) (29).

Diet quality was measured using the Healthy Eating Index 2010 (HEI-2010) (30). The HEI-2010 is a scoring system developed by the US Department of Agriculture that measures the degree of compliance to the 2010 Dietary Guidelines for Americans (31). Briefly, the HEI-2010 is made up of 12 food-based components: (i) total fruit; (ii) whole fruit; (iii) total vegetables; (iv) greens and beans; (v) whole grains; (vi) dairy; (vii) total protein foods; (viii) seafood and plant proteins; (ix) fatty acids; (x) refined grains; (xi) sodium and (xii) empty calories (defined as calories from solid fat, alcohol and added sugars). The first nine components represent adequacy components and the last three represents moderation components. The compliance to each of the 12 components is scored separately, and then summed together (scores ranging from 0 to 100) with higher scores indicating greater compliance with dietary recommendations.

Covariates

The covariates considered in the analyses were based on previous literature examining mediation analyses

between screen time and zBMI in adolescents (16). The covariates included age (in years), sex, ethnicity, socio-economic position (SEP), self-reported physical activity intensity and dietary intake under-reporting. SEP represents the family poverty-income-ratio and was calculated by dividing family income (reported by a parent) by the poverty guidelines (scores range 0–5), and self-reported physical activity intensity was calculated by averaging the metabolic equivalence score of all activities performed over the past 30 days (scores range 2.5–10). An objective accelerometer-derived measure of moderate-to-vigorous physical activity was initially considered as a covariate in the analyses; however, it reduced the sample by almost 50%. Dietary intake under-reporting was assessed on the basis of the ratio of total energy intake with estimated energy expenditure. Those with an energy intake to energy expenditure ratio of at least 2 standard deviations below the mean were classified as under-reporters (32).

Statistical analysis

To be included in the analyses (Figure 1), participants must be aged 12–19 years and not pregnant, have two days of complete dietary data obtained from the 24-h food recalls, have complete data on self-reported TV viewing, physical activity intensity, ethnicity and SEP, and have complete height and weight measures in order to calculate zBMI ($n=3,161$). To examine the presence of MetS, separate analyses were also undertaken from a subsample of participants who had the previous inclusion criteria as well as a fasting blood sample at the health examination visit ($n=1,379$).

Analyses were conducted using STATA/SE 14.0 software (StataCorp LP, College Station, Texas, 2015). To account for the complex survey sampling design, sample weights were applied to the descriptive statistics and to all mediation analyses. The 2-year weights were calculated to a combined 4-year weight by multiplying half of the assigned 2003–2004 and 2005–2006 2-year weight (e.g. $1/2 \times \text{WTDRD1}$). The 4-year dietary weight was then used for all analyses involving zBMI and the 4-year fasting blood sample weight was used in the analyses involving MetS. The significance level was set at $P < 0.05$ for all statistical tests.

Prior to the analyses, all variables were checked for normal distribution. Total energy intake, servings of fruit and vegetables, servings of discretionary snacks and servings of SSB were not normally distributed and were log transformed. Using the product of coefficients method by MacKinnon *et al.* (33), individual linear regression models were used to test whether each of the five dietary variables mediated the association between TV

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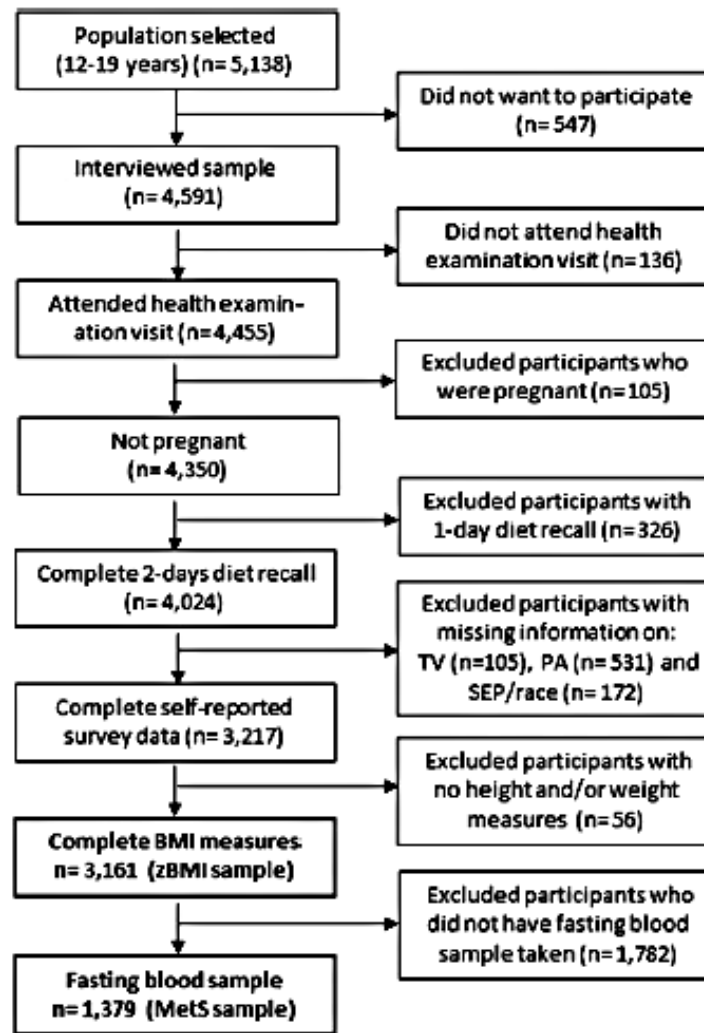


Figure 1 Study inclusion flow diagram of National Health and Nutrition Examination Survey 2003-2006 cohort and analytic sample.

viewing and zBMI or MetS (Figure 1). All dietary mediators were entered into separate regression models to minimize collinearity between variables (e.g. fruit and vegetables with dietary quality). All models controlled for age, sex, ethnicity, SEP, dietary intake under-reporting and self-reported physical activity.

As shown in Figure 2, for each mediation model, the following associations were tested: (i) associations between TV viewing time and the five dietary mediators (a-coefficient); (ii) associations between the five dietary mediators with zBMI or MetS, adjusting for TV viewing

time (b-coefficient); (iii) the total effect of TV viewing time and zBMI or MetS (c-coefficient) and (iv) the direct effect of TV viewing time with BMI or MetS, accounting for the dietary mediators (c'-coefficient). The mediating effect, also known as the indirect effect, of the dietary variables was calculated by multiplying the a and b coefficients ($a \times b$) and presented as a percentage of the mediated effect ($ab / (c' + ab)$). For the MetS outcome, the coefficients were used to calculate the mediating pathways; however, the odds ratios are presented in the tables for descriptive purposes.

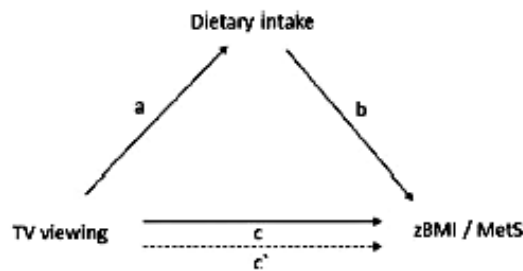


Figure 2 Theoretical diagram of the mediation pathways between TV viewing, dietary intake and zBMI/MetS. a-coefficient: association between TV viewing time with the dietary mediators; b-coefficient: association between the dietary mediators with zBMI/MetS; c-coefficient: total effect of TV viewing time with zBMI/MetS; c'-coefficient: the direct effect of TV viewing time with zBMI/MetS, accounting for the dietary mediators; zBMI: BMIz score, MetS: metabolic syndrome.

Results

Participant characteristics

Participant characteristics are shown in Table 1. Overall, 3,161 participants had complete zBMI profiles and 1,379 participants had complete data for identifying MetS. Approximately 53.1% of participants were male, and the mean age was 15.4 years. In the group with complete data for identifying MetS, 9.1% were classified as having MetS.

Relationship between TV viewing and diet (a-coefficient)

In the zBMI sample, TV viewing had a significant inverse association with diet quality, implying that for every hour of TV watched, diet quality decreased by 6.86 units (95%CI -1.45, -2.28) (Table 2). In the MetS sample, TV viewing was also inversely associated with dietary quality ($\beta = -7.59$; 95%CI -13.65, -1.53) and had a significant positive association with SSB consumption ($\beta = 0.39$; 95%CI 0.03, 0.76) (Table 3).

Relationship between diet and body mass index/metabolic syndrome (b-coefficient)

In the zBMI sample, total energy intake ($\beta = -0.09$; 95%CI -0.17, -0.00) and discretionary snacks ($\beta = -2.02$; 95%CI -3.16, -0.87) both showed a small inverse association with zBMI, after accounting for the covariates and TV viewing (Table 2). In the MetS sample, no significant associations were observed between any of the five dietary variables and MetS ($p > 0.05$) (Table 3).

Table 1 Demographic characteristics, blood profiles, TV viewing and dietary intake in US adolescents aged 12–19 years by health outcome (zBMI and MetS)

Characteristics	zBMI (n=3,161)	MetS (n=1,379)
Age, years	15.4 (2.2)	15.4 (2.3)
Sex [%], mean (95%CI)		
Boys	53.1 (50.8, 55.5)	53.1 (48.8, 57.5)
Girls	46.9 (44.5, 49.2)	46.9 (42.5, 51.2)
Ethnicity [%], mean (95%CI)		
Non-Hispanic white	65.9 (59.6, 71.7)	65.7 (59.4, 71.4)
Non-Hispanic black	13.7 (10.3, 18.1)	13.5 (10.3, 17.5)
Hispanic	10.6 (0.8, 14.5)	10.2 (7.3, 14.1)
Hispanic, other	3.4 (2.5, 4.8)	4.6 (3.2, 6.6)
Other race	6.3 (4.3, 9.1)	6.0 (3.8, 9.3)
SEP	2.7 (1.6)	2.7 (1.6)
Physical activity*	6.4 (1.4)	6.3 (1.4)
BMI		
kg/m ²	23.5 (5.6)	23.3 (5.7)
zBMI	0.6 (1.1)	0.5 (1.1)
Cardiometabolic		
Waist circumference, cm	n/a	81.1 (14.8)
Systolic BP, mm Hg	n/a	109.4 (10.3)
Diastolic BP, mm Hg	n/a	60.3 (11.5)
Plasma glucose, mmol/L	n/a	5.2 (1.0)
HDL-cholesterol, mg/dL	n/a	1.4 (0.3)
Triglycerides, mg/dL	n/a	1.0 (0.6)
MetS [%], mean (95% CI)	n/a	9.1 (8.7, 9.4)
TV viewing, h	2.2 (1.4)	2.1 (1.4)
Diet		
Total energy intake, kcal	2,248.0 (906.4)	2,219.1 (899.0)
Fruit/vegetables, servings	1.7 (1.1)	1.8 (1.3)
Discretionary snacks, servings	2.5 (2.1)	2.5 (2.2)
SSB, servings	2.1 (1.7)	2.1 (1.6)
Diet quality score, HEI-2010	40.8 (11.7)	40.8 (11.5)

Values weighted to account for survey design; mean and standard deviation in parentheses unless otherwise stated. SEP: socioeconomic position (range from 1 to 5); BMI, body mass index; zBMI: body mass index z score; BP: blood pressure; HDL: high-density lipoprotein; MetS: metabolic syndrome; SSB: sugar-sweetened beverages; n/a: sample does not have complete data.

*Physical activity intensity score (range from 2.5 to 10).

Relationship between TV viewing and body mass index/metabolic syndrome (c- and c'-coefficients)

Total

In the zBMI sample, TV viewing had a small but significant positive association with zBMI ($\beta = 0.99$; 95%CI 0.52, 1.46 $p < 0.001$), implying for every hour spent watching TV, zBMI increased by 0.99 units (equivalent to a 0.8 kg/m² increase in BMI) (Table 2). Similarly, in the MetS sample, TV viewing was also significantly related to having MetS, with for every hour spent watching

Table 2 Associations between TV viewing (h/day) with zBMI accounting for mediation by dietary variables (n = 3,161)

	a-coefficient	b-coefficient	c'-coefficient	ab	ab/c
	β (95% CI)	β (95% CI)	β (95% CI)	β (95% CI)	%
Total energy intake (kcal)	1.85 (-1.70, 5.41)	-0.09 (-0.17, -0.00)*	1.00 (0.53, 1.48)*	-0.16 (-0.50, 0.17)	-1.6
Fruit/vegetables (servings)	-0.14 (-0.34, 0.05)	-0.15 (-1.65, 1.35)	0.99 (0.51, 1.46)*	0.02 (-0.19, 0.23)	0.2
Discretionary snacks (servings)	0.16 (-0.17, 0.50)	-2.02 (-3.16, -0.87)*	1.02 (0.56, 1.48)*	-0.33 (-1.00, 0.34)	-3.3
SSB (servings)	0.15 (-0.18, 0.48)	0.74 (-0.58, 2.05)	0.98 (0.52, 1.44)*	0.11 (-0.19, 0.41)	1.1
Diet quality score (HEI-2010)	-6.86 (-11.45, -2.28)*	-0.01 (-0.07, 0.06)	0.98 (0.52, 1.45)*	0.05 (-0.36, 0.47)	0.5

Total effects (c-pathway) of TV viewing and zBMI: $\beta = 0.99$ (95%CI 0.52, 1.46) $p < 0.001$. Because of the small effect sizes, all results (except for the ab/c% column) have been multiplied by 10. Analyses adjusted for age, sex, ethnicity, socioeconomic position, self-reported physical activity intensity and dietary intake under-reporters. The unadjusted estimates are reported in Supplement file (Table S2). zBMI: body mass index z score; SSB: sugar-sweetened beverages.

* $P < 0.05$.

TV, the odds of having MetS increased by 18% (95% CI 1.00, 1.38; $p = 0.046$) (Table 3).

Direct

In the zBMI sample, similar to the total effect, but after accounting for each of the dietary mediators, the direct association between TV viewing and zBMI remained significant, and in some cases was stronger in magnitude ($p < 0.05$) (Table 2). For example, even after accounting for discretionary snacks, zBMI increased by 1.02 units (95%CI 0.56, 1.48) for every hour spent watching TV. However, in the MetS sample, the direct effect between TV and MetS was no longer significant after separately accounting for fruit and vegetable intake, SSB consumption and diet quality (Table 3).

Indirect/mediating

In the zBMI sample, when examining the indirect effect, or mediating effect, none of the five dietary variables showed a significant mediating effect on the relationship between TV viewing and zBMI (Table 2). However, when

examining the indirect effect in the MetS sample, SSB consumption and fruit and vegetable intake were both found to be significant positive mediators of the relationship between TV viewing and MetS, explaining 8.7% and 4.1% of the relationship respectively (Table 3). When examining the mediating effects of fruit and vegetable intake separately, only fruit intake remained a significant positive mediator explaining 5.2% of the relationship (results reported in Supplement file; Table S1).

Discussion

This study examined the relationships between TV viewing with BMI and MetS in a large representative sample of US adolescents, and whether the relationships were partially mediated by five elements of dietary intake. The findings suggest that TV viewing is positively associated with both BMI and MetS. After examining the mediating effects, no dietary mediators were observed in the TV viewing and zBMI relationship; however, SSB consumption and fruit and vegetable intake showed partial mediation effects in the TV and MetS relationship.

Table 3 Associations between TV viewing (h/day) with MetS accounting for mediation by dietary variables (n = 1,379)

	a-coefficient	b-coefficient	c'-coefficient	ab	ab/c
	β (95% CI)	OR (95% CI)	OR (95% CI)	β (95% CI)	%
Total energy intake (kcal)	-0.11 (-4.32, 4.09)	0.98 (0.94, 1.01)	1.17 (1.00, 1.38)*	0.03 (-6.58, 6.63)	0.2
Fruit/vegetables (servings)	-0.15 (-0.35, 0.06)	0.83 (0.30, 1.37)	1.17 (0.99, 1.37)	0.67 (0.29, 1.05)*	4.1*
Discretionary snacks (servings)	0.13 (-0.22, 0.47)	0.96 (0.69, 1.38)	1.18 (1.00, 1.33)*	-0.05 (-0.62, 0.52)	-0.3
SSB (servings)	0.39 (0.03, 0.76)*	1.44 (0.93, 2.22)	1.16 (0.98, 1.37)	1.43 (0.61, 2.25)*	8.7*
Diet quality score (HEI-2010)	-7.59 (-13.65, -1.53)*	0.99 (0.97, 1.00)	1.17 (0.99, 1.38)	0.87 (-0.37, 2.11)	5.3

Total effects (c-pathway) of TV viewing and MetS: OR 1.18 (95% CI 1.00, 1.38) $p = 0.046$. Because of the small effect sizes, the beta coefficient columns have been multiplied by 10. Analyses adjusted for age, sex, ethnicity, socioeconomic position, self-reported physical activity intensity and dietary intake under-reporters. The unadjusted estimates are reported in Supplement file (Table S3). MetS: metabolic syndrome, SSB: sugar-sweetened beverages.

* $P < 0.05$.

The finding that TV viewing was significantly associated with BMI is consistent with evidence from cross-sectional (7) and longitudinal studies (34), and reviews (8,35). Although, the literature on the TV viewing and MetS relationship in adolescents is limited, the findings from the present study are consistent with the few studies that have examined relationships between either TV viewing or screen time with markers of MetS or the presence of MetS (34,36,37). However, it is important to note, in both the current study and previous studies examining TV and MetS, the proportion of adolescents who were classified as having MetS was quite low (e.g. <10%) and results should be viewed with caution.

In addition, the finding that TV viewing was adversely associated with diet quality and positively associated SSB consumption is consistent with the literature. In an earlier National Health and Nutrition Examination Survey cohort examining TV viewing and dietary quality in 3,343 US adolescents, less time spent watching TV was associated with higher diet quality (13). Similarly, in a large cross-national study involving 162,305 adolescents from various countries, those who watched high amounts of TV were more likely to consume soft drink on a daily basis and less likely to consume fruit and vegetables (38).

However, the latter is in contrast to the current study where no significant associations were found between TV viewing with fruit and vegetable intake. In addition, other cross-sectional and longitudinal studies have reported positive associations between TV viewing and discretionary snacking (11,39), whereas the current study found no significant findings. These differences could be because of different definitions used to describe 'discretionary snacking' or different methods used to collect the dietary information (e.g. food frequency questionnaires vs. 24-h food recalls).

In the current study, many of the dietary elements were significantly related to BMI but not to MetS. Interestingly, two of the dietary elements, total energy intake and discretionary snacking, had counter-intuitive associations with BMI. For example, a lower energy intake and a lower intake of discretionary snacks were associated with a higher zBMI. These counter-intuitive associations were also observed in a previous mediation study (16) and could potentially be explained by social desirability bias where overweight or obese participants may under-report their dietary intake (40). The null findings between diet and MetS are in contrast to an earlier National Health and Nutrition Examination Survey study (1999–2002) examining MetS in adolescents. They found a higher intake of fruit and a higher diet quality score (measured via the HEI-2005) was associated with a decrease in MetS prevalence (15). The differences between studies could be because

of the low prevalence of MetS in both samples or to the different dietary quality scores that were used (e.g. HEI-2005 vs. HEI-2010).

Only one other cross-sectional study from Canada has examined the mediating effects of dietary intake on the relationship between TV and BMI in a large group of adolescents. Their results were similar to the current study, where no dietary variables were found to mediate the TV and BMI relationship (16). However, in a study involving younger children (aged 10 years), a combined unhealthy dietary variable consisting of high fat food, fruit juice and soda consumption was found to be a significant mediator in the TV viewing and BMI relationship (41). The differences in findings between studies may be because of different population groups being studied (children vs. adolescents). However, given the lack of studies that have performed mediation analyses, further studies examining both child and adolescent populations are needed to clarify whether any differences exist between these age groups.

To our knowledge, this is the first study to examine the mediating effects of dietary intake between TV and MetS. SSB consumption and fruit and vegetable intake were both found to partially mediate the TV and MetS relationship. SSB consumption is not a surprising mediator given the consistent associations it has with both TV viewing (42) and cardiometabolic health (14). However, the counter-intuitive findings that a higher intake of fruit and vegetables may mediate the TV and MetS relationship are difficult to explain. Similar to the counter-intuitive associations found between diet and BMI, social desirability bias may have occurred where participants with the MetS may have over-reported their intake of healthier food items such as fruit and vegetables (43,44). Alternatively, given this is a cross-sectional study, it is possible that some participants with MetS may have already been instructed to change their eating behaviours, and thus consume more fruit and vegetables.

The main strengths of this study include having a large representative-based sample and using objective measures of BMI status and MetS. Other strengths include the use of two 24-h recalls using the Automated Multiple-Pass Method to collect dietary intake data and adjusting for a variety of well-known confounders in the analyses. Limitations include the self-reported measures of TV viewing time, physical activity intensity and dietary intake as it is possible that under- or over-reporting may have occurred. Further, time spent in 'TV viewing' a decade ago may not be reflective of time spent in 'TV viewing' today. Advancing technology over the last decade has introduced watching TV on many portable devices (e.g. tablets, phones and computers); thus, 'total screen time' may be the contemporary equivalent term to use in

future studies assessing TV viewing. Additionally, using an objectively measured physical activity measure may have altered the results; however, by doing so it reduced the sample by almost 50%. Although a variety of covariates were adjusted for, other unmeasured covariates such as pubertal status may have influenced the observed associations. Further, given the number of mediation variables tested, multiple analyses were conducted for both the zBMI sample and MetS sample, which can lead to some findings being significant because of chance. Last, data on TV viewing and dietary intake were not collected for the same days and the cross-sectional design of the study limits the ability to determine the cause and effect relationship.

Conclusion

Contrary to our hypothesis, this study found that five aspects of dietary intake were not significant mediators in the TV viewing and BMI relationship. However, the consumption of SSB and fruit and vegetable intake were found to be significant mediators in the TV viewing and MetS relationship. This suggests that it is important to target TV viewing in interventions; however, the mechanisms explaining the health associations are still unclear. Further research examining other sedentary behaviours (e.g. computer use, video gaming), and total sedentary time is needed to determine if these behaviours have similar findings. Longitudinal and experimental studies should be conducted to better understand the role of dietary intake and how much, if any, it contributes to the TV viewing and cardiometabolic health relationship.

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Conflict of Interest Statement

The authors have no conflicts of interest.

Author contributions

The authors' responsibilities were as follows: EAF had full access to the data in the study and had primary responsibility for the integrity of the data, the accuracy of the data analysis, and the final content. EAF, SAM, KEL, DWD, JS: conceived and designed the study; EAF performed the statistical analysis with guidance from SAM, VC and JS. All authors were involved in writing the paper and had final approval of the submitted and published versions.

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Supporting Information

Additional supporting information may be found in the online version of this article at the publisher's web site.

Table S1: Associations between TV viewing (h/day) with MetS accounting for mediation effects of fruit and vegetable intake.

Table S2: Unadjusted associations between TV viewing (h/day) with zBMI accounting for mediation by dietary variables ($n = 3,161$).

Table S3: Unadjusted associations between TV viewing (h/day) with MetS accounting for mediation by dietary variables ($n = 1,379$).

4.3 Manuscript supplementary files

The supplementary files supporting the manuscript “*Mediating effects of dietary intake on associations of TV viewing, body mass index and metabolic syndrome in adolescents*” can be found on the following pages.

The supplementary files include:

- Table S1: Associations between TV viewing (hours/day) with metabolic syndrome accounting for mediation effects of fruit and vegetable intake
- Table S2: Unadjusted associations between TV viewing (hours/day) with zBMI accounting for mediating by dietary variables
- Table S3: Unadjusted associations between TV viewing (hours/day) with metabolic syndrome accounting for mediation by dietary variables.

Table S1: Associations between TV viewing (hours/day) with MetS accounting for mediation effects of fruit and vegetable intake

	a-coefficient B (95% CI)	b-coefficient OR (95% CI)	c'-coefficient OR (95% CI)	ab B (95% CI)	ab/c %
Fruit/Vegetables (servings)	-0.15 (-0.35, 0.06)	0.63 (0.30, 1.37)	1.17 (0.99, 1.37)	0.67 (0.29, 1.05)*	4.1*
Fruit only (servings)	-0.27 (-0.57, 0.04)	-0.31 (-1.11, 0.48)	0.16 (-0.01, 0.32)	0.85 (0.23, 1.46)*	5.2*
Vegetables only (serving)	0.02 (-0.20, 0.17)	-0.41 (-1.06, 0.25)	0.16 (0.00, 0.32)*	0.06 (-0.23, 0.35)	0.4

Total effects (c-pathway) of TV viewing and MetS: OR 1.18 (95% CI 1.00, 1.38) $p=0.046$.

Due to the small effect sizes, the beta coefficient columns have been multiplied by 10. Significance is noted by * $P < 0.05$

Analyses adjusted for age, sex, ethnicity, SEP, self-reported physical activity intensity and dietary intake under-reporters.

Table S2: Unadjusted associations between TV viewing (hours/day) with zBMI accounting for mediation by dietary variables (n=3,161)

	a-coefficient β (95% CI)	b-coefficient β (95% CI)	c'-coefficient β (95% CI)	ab β (95% CI)	ab/c %
Total energy intake (kcal)	1.24 (-3.29, 5.78)	-0.15 (-0.21,-0.09)*	1.17 (0.71, 1.64)*	-0.19 (-0.87, 0.48)	-1.7
Fruit/Vegetables (servings)	-0.16 (-0.35, 0.02)	-0.95 (-2.42, 0.52)	1.14 (0.72, 1.56)*	0.15 (-0.13, 0.44)	1.3
Discretionary snacks (servings)	0.05 (-0.26, 0.36)	-2.50 (-3.60, -1.39)*	1.17 (0.75, 1.59)*	-0.12 (-0.86, 0.61)	-1.1
SSB (servings)	0.17 (-0.23, 0.58)	-0.05 (-1.11, 1.02)	1.16 (0.74, 1.57)*	-0.01 (-0.19, 0.17)	-0.1
Diet quality score (HEI-2010)	-7.21 (-12.47, -1.95)*	0.01 (-0.05, 0.07)	1.16 (0.74, 1.58)*	-0.07 (-0.50, 0.35)	-0.6

Total effects (c-pathway) of TV viewing and zBMI: $\beta=1.16$ (95%CI 0.74, 1.57) $p<0.001$.

Due to the small effect sizes, all results (except for the ab/c% column) have been multiplied by 10. Significance is noted by * $P < 0.05$

zBMI: Body Mass Index z score; SSB: sugar-sweetened beverages

Table S3: Unadjusted associations between TV viewing (hours/day) with MetS accounting for mediation by dietary variables (n=1,379)

	a-coefficient	b-coefficient	c'-coefficient	ab	ab/c
	β (95% CI)	OR (95% CI)	OR (95% CI)	β (95% CI)	%
Total energy intake (kcal)	1.87 (-3.70, 7.45)	0.95 (0.92, 0.99)*	1.20 (1.04, 1.39)*	-0.88 (-10.92, 9.13)	-5.2
Fruit/Vegetables (servings)	-0.09 (-0.32, 0.13)	0.51 (0.25, 1.04)	1.18 (1.02, 1.37)*	0.62 (0.24, 0.99)*	3.6*
Discretionary snacks (servings)	0.10 (-0.24, 0.44)	0.76 (0.52, 1.10)	1.19 (1.03, 1.38)*	-0.28 (-0.86, 0.30)	-1.6
SSB (servings)	0.52 (0.14, 0.91)*	1.13 (0.69, 1.85)	1.18 (1.01, 1.38)*	0.64 (-0.36, 1.63)	3.8
Diet quality score (HEI-2010)	-8.62 (-14.68, -2.57)*	0.99 (0.98, 1.01)	1.18 (1.01, 1.37)*	0.62 (-0.89, 2.14)	3.7

Total effects (c-pathway) of TV viewing and MetS: OR 1.18 (95% CI 1.02, 1.37) $p=0.025$.

Due to the small effect sizes, the beta coefficient columns have been multiplied by 10. Significance is noted by * $P < 0.05$

MetS: Metabolic syndrome, SSB: sugar-sweetened beverages

4.4 Summary

Chapter Four examined the mediating role of five dietary variables (e.g. total energy intake, fruit and vegetable intake, consumption of discretionary snacks and sugar-sweetened beverages, and diet quality) of the relationships between TV viewing, BMI and metabolic syndrome in U.S. adolescents aged 12-19 years. Overall, positive associations were observed between TV viewing and BMI ($\beta= 0.99$, $p< 0.001$), and TV viewing and metabolic syndrome (OR= 1.18, $p= 0.046$). Contrary to the candidate's hypothesis, none of the dietary variables appeared to mediate the relationship between TV viewing and BMI. However, sugar-sweetened beverage consumption and fruit and vegetable intake partially mediated the relationship between TV viewing and metabolic syndrome, explaining 8.7% and 4.1% of the relationship, respectively.

These findings highlight the complex relationships observed between TV viewing, dietary intake and cardiometabolic health in adolescents. Further research is needed to better understand how much (if any) dietary intake contributes to the overall sedentary behaviour and cardiometabolic health relationship.

CHAPTER FIVE: STUDY TWO

MEDIATING EFFECTS OF DIET ON
ASSOCIATIONS OF VOLUME AND BOUTS OF
SEDENTARY TIME AND CARDIOMETABOLIC
HEALTH IN ADOLESCENTS

CHAPTER FIVE: STUDY TWO

MEDIATING EFFECTS OF DIET ON ASSOCIATIONS OF VOLUME AND BOUTS OF SEDENTARY TIME AND CARDIOMETABOLIC HEALTH IN ADOLESCENTS

5.1 Overview

Chapter Four examined the mediating effects of dietary intake on the relationships between TV viewing, BMI and metabolic syndrome in U.S. adolescents participating in the 2003-06 NHANES. Overall, dietary intake did not appear to mediate the TV viewing and BMI relationship and very few dietary variables were found to significantly mediate the relationship between TV viewing and metabolic syndrome. However, the mediating role of dietary intake when an objective measure of sedentary time (e.g. total volume and bouts of sedentary time) is examined with cardiometabolic outcomes is still unknown. Therefore, the following study will explore the mediating effects of dietary intake on the relationships between total volume and bouts of sedentary time with BMI and metabolic syndrome.

The following study has been published as a manuscript in *Obesity* (Impact factor; 3.614) and has been formatted according to their requirements. The citation for the manuscript is: *Fletcher EA, Carson V, McNaughton SA, Dunstan DW, Healy GN, Salmon J. Does diet mediate associations of volume and bouts of sedentary time with cardiometabolic health indicators in adolescents? Obesity. 2017; 25(3): 591-599.*

The manuscript supplementary files can be found directly after the manuscript in *Section 5.3*. The 'Authorship Statement' for this manuscript and a copy of the

‘Permission Statement to Publish’ this manuscript are contained in Appendix D.1 and Appendix D.2, respectively.

Findings from this manuscript were presented as a poster at the International Society of Behavioral Nutrition and Physical Activity conference in Cape Town, South Africa in June 2016 (poster available in Appendix D.3). The author contributions to the manuscript were as follows, with the candidate contributing to all aspects of the manuscript process: Fletcher, McNaughton, Dunstan and Salmon designed the study; Fletcher calculated the main variables used in the analyses with statistical guidance from Carson for the accelerometer data and McNaughton for the dietary data. Fletcher performed the main analyses with guidance from Carson and Healy; Fletcher wrote the initial draft of the manuscript; and Carson, McNaughton, Dunstan, Healy and Salmon provided critical edits and editions to the manuscript. All authors approved the final manuscript.

5.2 Manuscript

Original Article
PEDIATRIC OBESITY



Does Diet Mediate Associations of Volume and Bouts of Sedentary Time with Cardiometabolic Health Indicators in Adolescents?

Elly A. Fletcher¹, Valerie Carson², Sarah A. McNaughton¹, David W. Dunstan^{1,3,4,5,6,7}, Genevieve N. Healy^{3,5,8}, and Jo Salmon¹

Objective: Examine the mediating role of diet in the relationship between volume and duration of sedentary time with cardiometabolic health in adolescents.

Methods: Adolescents (12-19 years) participating in the 2003/04 and 2005/06 U.S. National Health and Nutrition Examination Survey (NHANES) were examined. Cardiometabolic health indicators were body mass index z-scores (zBMI) ($n = 1,797$) and metabolic syndrome (MetS) ($n = 812$). An ActiGraph hip-worn accelerometer was used to derive total sedentary time and usual sedentary bout duration. Dietary intake was assessed using two 24-hour dietary recalls. Mediation analyses were conducted to examine five dietary mediators [total energy intake, discretionary foods, sugar-sweetened beverages (SSB), fruits and vegetables, and dietary quality] of the relationship between total sedentary time and usual sedentary bout duration with zBMI and MetS.

Results: Total sedentary time was inversely associated with zBMI ($\beta = -1.33$; 95% CI -2.53 to -0.13) but attenuated after adjusting for moderate-to-vigorous physical activity. No significant associations were observed between usual sedentary bout duration with zBMI or either sedentary measure with MetS. None of the five dietary variables mediated any of the relationships examined.

Conclusions: Further studies are needed to explore associations of specific time periods (e.g., after school) and bout durations with both cardiometabolic health indicators and dietary behaviors.

Obesity (2017) 25, 591-599. doi:10.1002/oby.21750

Introduction

Adolescents with obesity and the metabolic syndrome (MetS) is a major public health concern in many Western countries (1). Over 20% of U.S. adolescents aged 12 to 19 years have obesity (2), and approximately 9.8% meet the criteria for MetS (3). Similar statistics have been reported in other Western countries such as Australia, Canada, and the UK (4). Since obesity tracks into adulthood (5), and adolescents who have MetS are at a higher risk of developing type 2 diabetes and cardiovascular disease during adulthood (6), it is

important to understand their lifestyle risk factors to help inform effective interventions.

Sedentary behavior, defined as any waking behaviors characterized by low energy expenditure (<1.5 metabolic equivalents) while in a sitting or reclining posture (7), has emerged as a health risk in the pediatric population. Evidence in adolescents suggests time spent engaged in certain sedentary behaviors, such as television viewing, is associated with overweight and obesity (8) and an increased risk

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Author contributions: EAF, SAM, DWD, and JS contributed to the design of the study; EAF performed the analyses and drafted the article; EAF, VC, SAM, DWD, GNH, and JS interpreted the data, revised it critically for important intellectual content, and approved the final version to be published.

Additional Supporting Information may be found in the online version of this article.

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of developing MetS (9). However, the few studies in adolescents examining accelerometry-measured total sedentary time with indicators of cardiometabolic health have reported mixed findings (10-12). In addition, evidence on the accumulation of sedentary time, for example, long “bouts” of sitting versus short “bouts” of sitting, also yields mixed findings (12-14).

These inconsistent findings in adolescents are in contrast to the evidence in the adult literature, both from experimental (15-17) and epidemiological studies (18,19), where prolonged, unbroken bouts of sedentary time have been found to be consistently associated with indicators of poor cardiometabolic health. These inconsistent findings pose the question as to whether sitting itself is a risk factor for poor cardiometabolic health in adolescents (e.g., whether sitting reduces skeletal muscle contractile activity and in turn reduces energy expenditure) or whether sitting is a *marker* of an unhealthy lifestyle. For example, due to the codependent nature of activities within a 24-hour period, time spent sedentary displaces time spent in more health enhancing activities, such as physical activity and sleep (20). Alternatively, time spent sitting could promote overeating or the consumption of unhealthy foods or beverages. For example, there is consistent evidence to show self-reported television viewing is associated with several unhealthy dietary behaviors (21); however, it is unclear whether dietary intake is also linked with objectively measured total sedentary time or bouts of sedentary time.

Given that the relationship between sedentary time and cardiometabolic health outcomes in adolescents is unclear, there is a need to better understand whether other factors, such as dietary intake, can help explain the inconsistent findings. One way to explore this is through mediation analyses, which allow the researcher to examine whether an underlying factor (e.g., mediator) may explain the pathway between an independent and dependent variable. Although some studies have explored the mediating pathway of dietary intake in the relationship between self-reported measures of sedentary behavior and BMI (22,23), to date no study has explored this relationship using objectively measured sedentary time. Therefore, this study aims to investigate whether five elements of dietary intake mediate the associations between both the volume and bouts of sedentary time with BMI and MetS in U.S. adolescents.

Methods

Study population and design

The National Health and Nutrition Examination Survey (NHANES) is a United States-based study that involves a multistage, stratified sampling design used to recruit a representative sample of the U.S. population (of all ages) in 2-year cycles. Further information about the NHANES study is described elsewhere (24); only data from adolescents (aged 12-19 years) collected in the 2003 to 2004 and 2005 to 2006 cycles were combined and considered for these analyses. Briefly, participants who were ≥ 18 years old provided consent and parents/guardians gave consent for minors (ages < 18 years). All eligible participants completed a questionnaire on their demographics and activity/sedentary behavior levels at a home interview. Afterward, participants (accompanied by their parent/guardian) were invited to attend a health examination visit to collect physiological measurements and complete one of two 24-hour dietary recalls. Those who attended the clinic were also asked to wear an activity

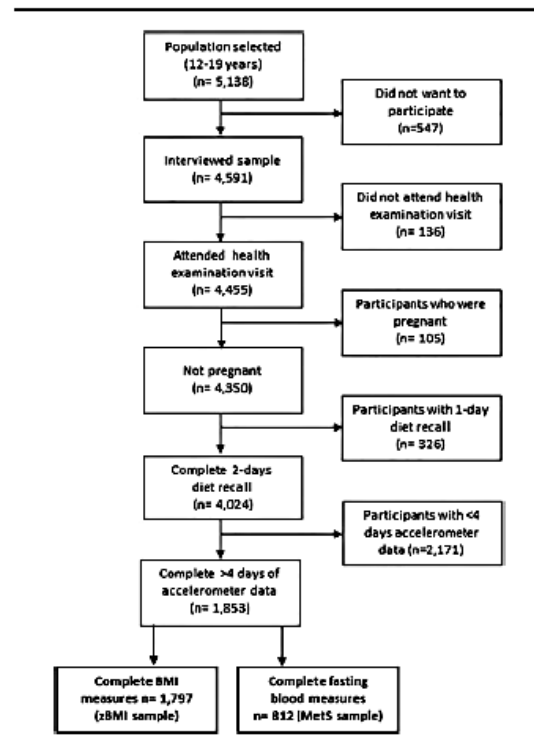


Figure 1 Participant flow diagram.

monitor for 7 days and undertake the second 24-hour dietary recall 4 to 11 days afterward. Participants who were not pregnant, had complete data on two 24-hour dietary recalls and anthropometric measurements, had 4 valid days of accelerometry data, and had complete fasting blood samples were included in the analyses (Figure 1). Overall, 35% and 16% of participants from the original cohort had complete data for the BMI sample and MetS sample, respectively.

Measurements

Cardiometabolic measurements. Height, weight, and waist circumference measurements and blood pressure were taken from all participants who attended the health examination visit. Standing height (centimeters) was measured without shoes using a stadiometer and weight (kilograms) was measured in light clothing using digital scales. Height and weight were used to calculate participants' zBMI using age- and sex-BMI percentiles based on the 2000 Centers for Disease Control and Prevention U.S. growth charts (25). Fasting blood glucose and serum lipids were obtained from a subsample of participants who attended the morning session and had fasted overnight for at least 8.5 hours. The full protocol and assessment on blood analyses can be found elsewhere (26). The presence of MetS

was calculated using the International Diabetes Federation criteria, specifically designed for the pediatric population (27). For these analyses, adolescents were considered to have MetS if they had an elevated waist circumference according to age- and sex-specific percentiles (≥ 79.5 to ≥ 102.0 cm) and had two or more of the following four risk factors (cutoff values presented are dependent on age and sex): elevated systolic (≥ 121 to ≥ 130 mm Hg) or diastolic blood pressure (≥ 76 to ≥ 85 mm Hg), low HDL-cholesterol (≤ 1.13 to ≤ 1.3 mmol/L), elevated triglycerides (≥ 1.44 to ≥ 1.70 mmol/L), and elevated plasma glucose (≥ 5.6 mmol/L).

Sedentary time. Sedentary time was measured via an ActiGraph AM-7164 accelerometer (LLC, Fort Walton Beach, FL), an accurate and reliable tool that quantifies sedentary behavior and moderate-to-vigorous physical activity (MVPA) (28). Participants wore the monitor on their right hip during all waking hours for 7 days, except for bathing and swimming. Data were downloaded in 1-minute epochs. Non-wear time was defined as at least 20 minutes of zero counts. Daily sedentary time was defined as all wear-time minutes with an average activity count of ≤ 100 cpm. Sedentary time was standardized for wear time using the residual method (29). Usual sedentary bout was calculated using the equation by Chastin and Granat (30). Briefly, a sedentary bout was calculated by summing all uninterrupted minutes ≤ 100 cpm. Taking into account different bout durations, the midpoint of all the sedentary bouts that lie on the sedentary accumulation curve was then calculated for each participant. Analyses were limited to participants who had ≥ 10 hours of monitor wear time on any ≥ 4 days, and the monitor was returned in calibration.

Dietary intake. Dietary intake was assessed via two interviewer-administered 24-hour dietary recalls, delivered via a computer-assisted system (the Automated Multi-Pass Method). Participants aged 12 years and older reported their first dietary recall with an interviewer at the health examination visit and then 4 to 11 days later by telephone. All dietary data were coded using the USDA National Nutrient Database for Standard Reference (31). Based on previous literature examining dietary mediators (22), the following dietary variables were examined: (1) total energy intake; (2) consumption of discretionary foods; (3) consumption of sugar-sweetened beverages (SSB); (4) fruit and vegetable intake; and (5) diet quality.

All dietary variables were calculated based on the average of 2 days of dietary recall. Total energy intake (calories) was calculated for each participant based on the quantity of food and beverages reported, and servings of discretionary foods, SSB, and fruits and vegetables were calculated using the Food Patterns Equivalents Database (32). Discretionary foods were defined as grain- and dairy-based desserts, cereal and granola bars, sweet snacks and candies, sugar-syrups and preserves, salty snacks from grain or starchy vegetables, and dips/spreads. SSB were defined as any nonalcoholic beverage with added sugar including soda, fruit-flavored drinks (not 100% juice), sweetened tea, coffee and milk drinks, sport drinks, and energy drinks. Any "diet" drinks, 100% fruit juice, or unsweetened tea or coffee were not included as a SSB. A serving of discretionary foods and a serving of SSB were equivalent to 143 calories (600 kJ) (33).

Fruit and vegetable intake included any whole fruit (not including fruit juice), and vegetables included potatoes, beans, and legumes. A

serving of fruits and vegetables was equivalent to one cup of raw, canned, or frozen fruits or vegetables or two cups of leafy green vegetables. Diet quality was calculated using The Healthy Eating Index 2010 (HEI-2010) (34), a scoring system that measures the degree of compliance to the 2010 Dietary Guidelines for Americans. The HEI-2010 is made up of 12 food-based components and compliance to each of the 12 components was scored separately and then summed together. Scores ranged from 0 to 100, and higher scores indicated greater compliance to the dietary recommendations.

Covariates. The covariates considered for the analyses were age (in years), sex, race/ethnicity, socioeconomic position, dietary intake under-reporting, and objectively measured MVPA. For the nonenergy-related mediators, total energy intake was also adjusted for in the main analyses. Race/ethnicity was categorized as Mexican American, other Hispanic, non-Hispanic white, non-Hispanic black, and other race, including multiracial. Socioeconomic position was calculated by dividing family income with the poverty guidelines specific to family size, year, and state. Dietary intake under-reporting was assessed on the basis of the ratio of total energy intake with estimated energy expenditure. Those below the energy intake to energy expenditure ratio of 2 standard deviations or more were classified as under-reporters (35). MVPA was calculated according to the Freedson accelerometer age-cut points for adolescents (36).

Statistical analysis

Analyses were conducted using SAS[®] 9.4 (SAS Institute, Inc., Cary, NC) and STATA/SE[™] 14.0 software (StataCorp LP) using survey commands. To obtain population-representative findings, regression estimates with weightings were applied to the descriptive statistics and to all mediation analyses. As recommended in the NHANES Analytic Guidelines, 2-year sample weights for each NHANES cycle (2003 to 2004 and 2005 to 2006) were combined to provide 4-year sample weights. Given $>10\%$ of participants had missing data (mostly due to accelerometer noncompliance), the sample weights were recalculated using age, sex, and race/ethnicity. The day 2 dietary sample weights were used for the main analyses involving zBMI and the fasting subsample weights were used for the analyses involving MetS. The significance level was set at $P < 0.05$ for all statistical tests.

Prior to the main analyses, total energy intake, servings of discretionary foods, SSB, and fruits and vegetables, and usual sedentary bout were not normally distributed so were log transformed. Using the product of coefficients method by MacKinnon et al. (37), regression analyses were used to test whether each dietary variable mediated the association between sedentary time (total and usual bout) with zBMI and MetS (Figure 2). Based on recent evidence on the compositional paradigm for codependent data (20), the first analyses (Model 1) adjusted for age, sex, ethnicity, socioeconomic position, and dietary intake under-reporters, and the second analyses (Model 2) additionally adjusted for MVPA (Model 2). For both models, the nonenergy-related additionally adjusted for total energy intake in the main analyses.

As shown in Figure 2, for each mediation model, the following associations were tested: (1) associations of sedentary time (total and usual bout) with each of the five dietary mediators (a-coefficient pathway); (2) associations of the five dietary mediators with zBMI or MetS, adjusting for either total sedentary time or usual sedentary

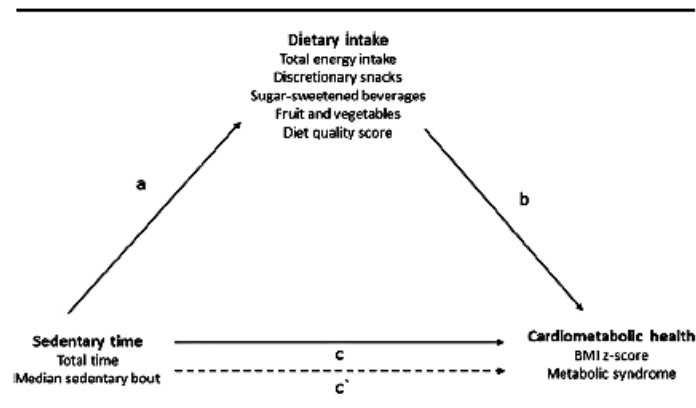


Figure 2 Theoretical diagram of the mediation pathways between sedentary time, dietary intake, and cardiometabolic health.

bout (b-coefficient pathway); (3) associations of sedentary time (total and usual bout) with zBMI or MetS (c-coefficient pathway); and (4) the direct association of sedentary time (total and usual bout) with zBMI or MetS, accounting for each of the dietary mediators (c'-coefficient pathway). The mediating effect of the dietary variables was calculated by multiplying the a- and b-coefficients ($a \times b$) (37). As MetS is a dichotomous outcome, the coefficients were used to calculate the mediating pathways; however, the odds ratios (OR) are presented in the tables for descriptive purposes.

Results

The baseline characteristics of those adolescents with complete zBMI profiles ($n = 1,797$) and complete data for identifying MetS ($n = 812$) are presented in Table 1. Compared with those excluded in the analyses ($n = 2,643$), participants included in the current analyses ($n = 1,797$) were significantly younger, included more Mexican American participants and fewer non-Hispanic white participants, and had a higher socioeconomic position, lower BMI, lower usual sedentary bout duration, higher fruit and vegetable intake and HEI score, a higher consumption of discretionary foods, and a lower consumption of SSB (Supporting Information Table S1).

Sedentary time and dietary intake

In the zBMI sample, total sedentary time was positively associated with SSB consumption ($\beta = 0.67$; 95% CI 0.01 to 1.33) after adjusting for age, sex, ethnicity, socioeconomic position, dietary intake under-reporting, and MVPA, with no other significant associations observed for usual sedentary bout with any of the dietary variables (Table 2). In the MetS sample, no associations were observed for total sedentary time and usual sedentary bout with any of the dietary variables ($P > 0.05$) (Table 3).

Dietary intake, zBMI, and MetS

In the zBMI sample, total energy intake was inversely associated with zBMI ($\beta = -0.17$; 95% CI -0.30 to -0.03) after separately

taking into account total sedentary time and usual sedentary bout and remained significant after adjusting for MVPA ($\beta = -0.16$; 95% CI -0.29 to -0.03). Consumption of discretionary foods was also inversely associated with zBMI after adjusting for MVPA only ($\beta = -1.76$; 95% CI -3.16 to -0.36) (Table 2). In the MetS sample, a higher total energy intake was significantly associated with a reduction in the risk of having MetS by 12%, after separately accounting for total sedentary time and usual sedentary bout duration. The findings remained significant after adjusting for MVPA (Table 3).

Sedentary time, zBMI, and MetS

After adjusting for age, sex, ethnicity, socioeconomic position, and dietary intake under-reporting, the total effect (e.g., c-coefficient) between total sedentary time and zBMI was $\beta = -1.33$ (CI -2.53 to -0.13), implying that for every hour spent sitting, zBMI decreased by 1.33 units (Table 2). However, after additionally adjusting for MVPA, no significant association remained. When examining the total effect between sedentary time and MetS, total sedentary time or usual sedentary bout was not significantly associated with having MetS, even without accounting for MVPA (Table 3).

When examining the direct effect (c'-coefficient) of sedentary time with zBMI, an inverse relationship was observed between total sedentary time and zBMI when accounting for each of the dietary variables (Table 2). However, after adjusting for MVPA, no significant associations remained. When examining usual sedentary bout with zBMI, there were no significant associations when accounting for each of the dietary variables and MVPA ($P > 0.05$). In the MetS sample, no direct effect was observed between sedentary time or usual sedentary bout with MetS after separately accounting for all of the dietary variables (Table 3). When examining the indirect effect (e.g., mediation), none of the five dietary variables examined was found to have a significant mediation effect on the associations between sedentary time (total or bout duration) with zBMI or MetS ($P > 0.05$).

TABLE 1 Demographic characteristics, blood profiles, sedentary/activity time, and dietary intake in U.S. adolescents participating in NHANES, 2003-2006

Variable	zBMI (n = 1,797)		MetS (n = 812)	
	Mean	95% CI	Mean (%) ^a	95% CI
Age (y)	15.1	14.9-15.3	15.1	14.9-15.4
Sex (%)				
Boys	51.1	47.7-54.5	50.1	46.0-55.7
Girls	48.9	45.5-52.3	49.1	44.3-54.0
Ethnicity (%)				
Mexican American	34.4	32.2-36.6	35.2	32.0-38.6
Other Hispanic	3.2	2.5-4.1	3.1	2.1-4.5
Non-Hispanic white	23.8	21.9-25.8	21.7	19.0-24.6
Non-Hispanic black	34.4	32.3-36.7	35.5	32.2-38.8
Other race	4.2	3.3-5.2	4.6	3.3-6.2
Socioeconomic position	2.7	2.5-2.9	2.7	2.4-2.9
BMI				
kg/m ²	22.8	22.3-23.3	22.8	22.1-23.3
z-score	0.5	0.4-0.7	0.5	0.3-0.6
Overweight (%)	23.5	18.9-28.8	20.3	16.2-25.2
Obesity (%)	12.1	9.4-15.4	13.9	10.5-18.2
Cardiometabolic components				
Waist circumference (cm)			79.2 (34.1)	77.7-80.7
Systolic BP (mm Hg)			108.5 (4.8)	107.2-109.7
Diastolic BP (mm Hg)			60.6 (1.1)	59.4-61.8
Plasma glucose (mg/dL)			5.1 (9.6)	5.0-5.2
HDL-cholesterol (mg/dL)			1.4 (22.2)	1.3-1.4
Triglycerides (mg/dL)			0.9 (11.4)	0.9-1.0
MetS (%)			6.7	4.2-10.5
Sedentary time				
Total (h)	6.9	6.8-6.9	7.1	7.0-7.2
Usual bout (min)	8.8	8.4-9.1	9.3	8.9-9.8
MVPA (min)	26.1	23.4-28.8	26.3	22.7-29.8
Diet				
Total energy intake (kcal)	2,224	2150-2298	2241.4	2180-2301
Discretionary foods (servings)	2.6	2.4-2.9	2.7	2.4-2.9
SSB (servings)	2.0	1.8-2.1	1.9	1.8-2.1
Fruits/vegetables (servings)	1.8	1.7-1.9	1.8	1.7-2.0
Diet quality score (HEI-2010)	41.8	40.4-43.2	41.5	40.1-42.9

^a% of those meeting the individual components of MetS in parentheses. Values weighted to account for survey design. Mean and 95% CI. BP, blood pressure; HDL, high-density lipoprotein; HEI-2010, Healthy Eating Index 2010; MetS, metabolic syndrome; MVPA, moderate-to-vigorous physical activity; SSB, sugar-sweetened beverages.

Discussion

This study examined the mediating effects of five dietary variables in the relationship between total sedentary time and usual sedentary bout with zBMI and MetS. Overall, total sedentary time was inversely associated with zBMI; however, the association was no longer significant after adjusting for MVPA. No significant associations were observed between usual sedentary bout and zBMI or between total sedentary time and usual sedentary bout with MetS. Although some of the dietary variables were independently related to total sedentary time, zBMI, and MetS, none of the dietary variables was a significant mediator.

The counterintuitive finding between sedentary time and zBMI when not accounting for MVPA is in contrast to other cross-sectional studies in adolescents that found positive associations between objectively measured sedentary time with insulin resistance (12), fasting glucose, triglycerides, blood pressure, and a cardiovascular risk score (9). However, it is important to note, these two studies did not adjust for MVPA in the analyses. When this study adjusted for MVPA, the results between total sedentary time and zBMI attenuated. This has also been observed in other studies that examined the unadjusted and adjusted results of MVPA in the analyses and found the significant associations between objectively measured sedentary time and cardiometabolic components also

TABLE 2 Associations between sedentary time (total and usual sedentary bout) with zBMI, accounting for mediation by dietary variables (n = 1,797)

Outcome: zBMI	a-Coefficient β (95% CI)		b-Coefficient β (95% CI)		c-Coefficient β (95% CI)		ab (mediated) β (95% CI)	
	Model 1	Model 2	Model 1	Model 2	Model 1	Model 2	Model 1	Model 2
Total sedentary time (h)								
Mediators								
Total energy intake (kcal/d) ^a	7.05 (-2.94 to 17.05)	5.88 (-3.63 to 15.39)	-0.17 (-0.30 to -0.03) ^b	-0.16 (-0.29 to -0.03) ^b	-1.21 (-2.40 to -0.03) ^b	-1.02 (-2.24 to 0.19)	-1.16 (-3.00 to 0.67)	-0.94 (-2.57 to 0.70)
Discretionary foods (servings/d) ^a	-0.11 (-1.04 to 0.81)	-0.32 (-1.22 to 0.58)	-1.87 (-3.23 to -0.50)	-1.76 (-3.16 to -0.36) ^b	-1.23 (-2.41 to -0.04) ^b	-1.08 (-2.30 to 0.15)	0.21 (-1.46 to 1.89)	0.56 (-1.02 to 2.14)
SSB (servings/d) ^a	0.59 (-0.10 to 1.29)	0.67 (0.01 to 1.33) ^b	0.55 (-1.07 to 2.17)	0.48 (-1.09 to 2.05)	-1.24 (-2.43 to -0.04) ^b	-1.05 (-2.27 to 0.16)	0.33 (-0.67 to 1.32)	-0.32 (-0.73 to 1.38)
Fruits and vegetables (servings/d) ^a	-0.32 (-0.85 to 0.21)	-0.39 (-0.91 to 0.13)	-0.39 (-2.37 to 1.58)	-0.29 (-2.30 to 1.73)	-1.22 (-2.49 to -0.04) ^b	-1.03 (-2.25 to 0.18)	0.13 (-0.52 to 0.77)	0.11 (-0.65 to 0.87)
Diet quality score (HEI-2010)	-3.47 (-17.32 to 10.38)	-4.93 (-17.95 to 8.09)	-0.05 (-0.53 to 0.85)	-0.04 (-0.13 to 0.04)	-1.35 (-2.54 to -0.15) ^b	-1.14 (-2.35 to 0.07)	0.16 (-0.53 to 0.85)	0.22 (-0.47 to 0.90)
Usual sedentary bout (min)^a								
Mediators								
Total energy intake (kcal/d) ^a	1.05 (-9.68 to 11.78)	2.06 (-8.71 to 12.83)	-0.17 (-0.31 to -0.04) ^b	-0.16 (-0.30 to -0.03) ^b	-1.12 (-2.42 to 0.18)	-1.30 (-2.65 to 0.06)	-0.18 (-1.97 to 1.60)	-0.34 (-2.05 to 1.37)
Discretionary foods (servings/d) ^a	0.38 (-0.64 to 1.40)	0.53 (-0.58 to 1.63)	-1.82 (-3.21 to -0.44) ^b	-1.68 (-3.11 to -0.26) ^b	-1.06 (-2.37 to 0.24)	-1.22 (-2.57 to 0.13)	-0.69 (-2.55 to 1.16)	-0.88 (-2.81 to 1.04)
SSB (servings/d) ^a	-0.53 (-1.30 to 0.24)	-0.58 (-1.37 to 0.22)	0.37 (-1.25 to 2.00)	0.30 (-1.28 to 1.89)	-1.11 (-2.43 to 0.21)	-1.29 (-2.66 to 0.08)	-0.20 (-1.07 to 0.68)	-0.17 (-1.08 to 0.73)
Fruits and vegetables (servings/d) ^a	-0.47 (-1.21 to 0.28)	-0.44 (-1.16 to 0.29)	-0.35 (-2.38 to 1.67)	-2.26 (-2.30 to 1.79)	-1.15 (-2.46 to 0.16)	-1.32 (-2.67 to 0.03)	0.17 (-0.78 to 1.11)	0.11 (-0.76 to 0.98)
Diet quality score (HEI-2010)	8.84 (-13.92 to 31.61)	9.86 (-12.46 to 32.18)	-0.04 (-0.13 to 0.05)	-0.04 (-0.12 to 0.05)	-1.10 (-2.41 to 0.21)	-1.29 (-2.65 to 0.07)	-0.38 (-1.59 to 0.82)	-0.39 (-1.56 to 0.78)

Model 1: Total effects (c-pathway) of total sedentary time $\beta = -1.33$ (95% CI -2.53 to -0.13)^b and usual sedentary bout $\beta = -1.14$ (95% CI -2.44 to 0.17) with zBMI. Analyses adjusted for age, sex, ethnicity, socioeconomic position, and dietary intake under-reporting.
 Model 2: Total effects (c-pathway) of total sedentary time $\beta = -1.12$ (95% CI -2.33 to 0.09) and usual sedentary bout $\beta = -1.33$ (95% CI -2.68 to 0.02) with zBMI. Analyses adjusted for age, sex, ethnicity, socioeconomic position, dietary intake under-reporting, and moderate-to-vigorous physical activity. For both models, the nonenergy-related mediators additionally adjusted for total energy intake in the main analyses. Due to the small units of measure, all results (except for the ab/c% column) have been multiplied by 10.
^aLog transformed.
^bSignificance is noted by $P < 0.05$.
 HEI-2010, Healthy Eating Index 2010; SSB, sugar-sweetened beverages.

TABLE 3 Associations between sedentary time (total and usual sedentary bout) with MeS, accounting for mediation by dietary variables (n = 812)

Outcome: MeS	a-Coefficient β (95% CI)		b-Coefficient OR (95% CI)		c-Coefficient OR (95% CI)		ab (mediated) β (95% CI)	
	Model 1	Model 2	Model 1	Model 2	Model 1	Model 2	Model 1	Model 2
Total sedentary time (h)								
Mediators								
Total energy intake (kcal/d) ^a	9.18 (-6.12 to 24.48)	7.92 (-7.99 to 23.82)	0.88 (0.81 to 0.95) ^b	0.88 (0.81 to 0.95) ^b	0.73 (0.20 to 2.65)	0.66 (0.21 to 2.04)	-12.01 (32.50 to 8.49)	-10.37 (-31.30 to 10.57)
Discretionary foods (servings/d) ^a	-1.07 (-2.42 to 0.28)	-1.16 (-2.59 to 0.27)	0.85 (0.38 to 1.88)	0.82 (0.37 to 1.81)	0.71 (0.20 to 2.50)	0.64 (0.21 to 1.87)	1.75 (-6.67 to 10.17)	2.29 (-6.93 to 11.51)
SSB (servings/d) ^a	0.15 (-0.85 to 1.52)	0.20 (-0.95 to 1.35)	1.05 (0.55 to 2.02)	1.10 (0.55 to 2.20)	0.73 (0.20 to 2.63)	0.66 (0.22 to 2.00)	0.08 (-1.03 to 1.19)	0.20 (-1.52 to 1.91)
Fruits and vegetables (servings/d) ^a	-0.20 (-0.84 to 0.43)	-0.19 (-0.91 to 0.05)	0.71 (0.20 to 2.60)	0.70 (0.19 to 2.52)	0.72 (0.20 to 2.61)	0.65 (0.21 to 2.00)	0.69 (-2.56 to 3.94)	0.69 (-2.74 to 4.11)
Diet quality score (HEI-2010)	-7.61 (-30.41 to 15.19)	-8.20 (-31.77 to 15.37)	1.01 (0.97 to 1.04)	1.00 (0.98 to 1.00)	0.69 (0.21 to 2.26)	0.64 (0.22 to 1.83)	-0.42 (-3.42 to 2.59)	-0.48 (-3.68 to 2.73)
Usual sedentary bout (min)^a								
Mediators								
Total energy intake (kcal/d) ^a	-5.55 (-21.43 to 10.33)	-5.10 (-20.63 to 10.43)	0.88 (0.81 to 0.95) ^b	0.88 (0.81 to 0.95) ^b	1.78 (0.75 to 4.21)	1.89 (0.74 to 4.81)	7.34 (-13.26 to 27.94)	6.70 (-13.31 to 26.72)
Discretionary foods (servings/d) ^a	0.68 (-0.22 to 1.59)	0.70 (-0.22 to 1.63)	0.85 (0.39 to 1.85)	0.83 (0.38 to 1.80)	1.81 (0.77 to 4.27)	1.94 (0.76 to 4.92)	-1.13 (-6.44 to 4.18)	-1.35 (-6.85 to 4.16)
SSB (servings/d) ^a	-0.98 (-2.07 to 0.20)	-0.95 (-2.09 to 0.19)	1.18 (0.65 to 2.16)	1.23 (0.69 to 2.20)	1.83 (0.77 to 4.33)	1.94 (0.76 to 4.92)	-1.57 (-7.26 to 4.12)	-1.96 (7.72 to 3.80)
Fruits and vegetables (servings/d) ^a	-0.30 (-1.26 to 0.66)	-0.31 (-1.24 to 0.62)	0.79 (0.26 to 2.69)	0.80 (0.24 to 2.68)	1.76 (0.76 to 4.10)	1.88 (0.75 to 4.70)	0.70 (-3.62 to 5.02)	0.71 (-3.44 to 4.85)
Diet quality score (HEI-2010)	16.29 (-11.42 to 44.00)	16.45 (-11.60 to 44.50)	1.00 (0.97 to 1.01)	1.00 (0.99 to 1.00)	1.77 (0.72 to 4.35)	1.91 (0.72 to 5.07)	0.51 (-4.81 to 5.82)	0.50 (-4.65 to 5.65)

Model 1: Total effects (c-pathway) of total sedentary time OR = 0.64 (95% CI 0.22 to 1.84) and usual sedentary bout OR = 1.92 (95% CI 0.71 to 5.16) with MeS. Analyses adjusted for age, sex, ethnicity, socioeconomic position, and dietary intake under-reporting.
 Model 2: Total effects (c-pathway) of total sedentary time OR = 0.64 (95% CI 0.22 to 1.84) and usual sedentary bout OR = 1.92 (95% CI 0.71 to 5.16) with MeS. Analyses adjusted for age, sex, ethnicity, socioeconomic position, dietary intake under-reporting, and moderate-to-vigorous physical activity. For both models, the nonenergy-related mediators additionally adjusted for total energy intake in the main analyses. Due to the small units of measure, the a-coefficient and ab-coefficient have been multiplied by 10.
^aLog transformed.
^bSignificance is noted by P < 0.05.
 HEI-2010, Healthy Eating Index 2010; SSB, sugar-sweetened beverages.

attenuated (10,14). On the other hand, high levels of MVPA (i.e., approximately 60-75 min/d) have recently been shown to eliminate the increased risk of all-cause mortality associated with high amounts of sedentary time (38). To build upon the current evidence to date (20), further research should utilize an integrated paradigm to understand the collective health implications of sedentary behavior and MVPA on pediatric obesity and MetS, rather than examining the effects in isolation.

In this study, total sedentary time was only positively associated with SSB consumption and not with any other dietary variables. The study found for every hour spent sedentary, SSB intake increased by 0.67 servings. Although this association is small, this is equivalent to consuming 14 extra calories each day which in turn can increase weight by 0.6 kg within 1 year (39). The lack of association between total sedentary time and the other dietary elements is in contrast to the literature examining television viewing, where time spent watching TV has consistently been shown to be related to unhealthy dietary elements, such as a higher intake of energy-dense snacks, fast food, and total energy intake in youth (21). The stronger links between television viewing and unhealthy dietary habits are thought to be due to advertising where foods high in fat and sugar are frequently advertised on children's television programs (40). Thus, it appears that specific sedentary behaviors like television viewing have stronger links with dietary intake as opposed to the total time spent being sedentary or accumulating bouts of sedentary time.

When examining the association between dietary intake and cardiometabolic health, this study found an inverse relationship between total energy intake with both zBMI and MetS and between discretionary snacking and zBMI. Although the findings were not in the expected direction, it is possible that participants with a larger zBMI or those at risk of having MetS may have already sought professional help and thus started to decrease their energy intake or discretionary snacking.

Although one study in adolescents found diet to partially mediate the relationship between television viewing with zBMI (23), this study found none of the dietary variables significantly mediated the relationship between objectively measured sedentary time and bouts with either zBMI or MetS. This could be simply due to the sedentary time not being matched with the dietary intake. For example, the 2 × 24 hour dietary recalls were performed on different days, not necessarily aligning with when the accelerometers were worn to capture sedentary time. This makes it difficult to assess how much of the sedentary time spent throughout the day is also engaging in an eating occasion. Future studies are needed to examine both sedentary time and eating occasions concurrently, and whether adolescents who engage in high amounts of sedentary time and a high number of eating occasions are at a higher risk of adverse cardiometabolic health than those who engage in the same amount of high sedentary time but have fewer eating occasions.

Strengths and limitations

Strengths of this study include the objective measures of sedentary time and health outcomes and the use of 2 × 24 hour dietary recalls allowing for investigation of individual dietary behaviors and overall diet quality. This study also adjusted for a variety of well-known covariates, including dietary intake under-reporting, which would have reduced the measurement error associated with self-reporting

dietary intake but would not have completely eliminated it. Limitations include the cross-sectional design of the study which cannot distinguish between cause and effect and the MacKinnon method used for the analyses which does not account for multiple comparisons. Additionally, although the accelerometer provides an objective measure of sedentary time, accelerometers cannot assess contextual factors which make it difficult to assess what the participants were doing while sedentary and whether they were consuming food and beverages. Lastly, only a small percentage of participants were classified as having MetS; thus, this may not have been large enough to see associations.

Conclusion

In conclusion, this study found objectively measured sedentary time was associated with zBMI; however, the association attenuated after accounting for MVPA. The study also found that total sedentary time was not associated with MetS, nor was the usual sedentary bout duration with either zBMI or MetS. Although the study found some associations between dietary intake with the health outcomes, none of the dietary variables was a significant mediator in the sedentary time, zBMI, and MetS relationship. Given the lack of associations found when examining total sedentary time and bouts of sedentary time, this suggests that intervention programs may have to address certain sedentary behaviors (i.e., television viewing) differently than total sedentary time. **○**

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5.3 Manuscript supplementary files

The supplementary file supporting the manuscript “*Does diet mediate associations of volume and bouts of sedentary time with cardiometabolic health indicators in adolescents?*” can be found on the following page.

The supplementary file includes:

- Table S1: Comparison between U.S. adolescents included in the zBMI analyses versus those excluded.

Table S1: Comparison between U.S. adolescents included in the zBMI analyses (n=1,797) versus those excluded (n=2,643)

Variables	Included (n= 1,797)		Excluded (n=2,643)	
Age (years)	15.1	14.9, 15.3*	15.8	15.7, 15.9
Sex (%)				
Boys	51.1	47.7, 54.5	50.2	48.6, 52.3
Girls	48.9	45.5, 52.3	49.8	47.7, 51.9
Ethnicity (%)				
Mexican American	34.4	32.2, 36.6*	29.7	27.9, 31.4
Other Hispanic	3.2	2.5, 4.1	2.8	2.3, 3.6
Non-Hispanic white	23.8	21.9, 25.8*	28.0	26.4, 29.8
Non-Hispanic black	34.4	32.3, 36.7	35.1	33.3, 36.9
Other race	4.2	3.3, 5.2	4.2	3.5, 5.0
Socioeconomic position	2.7	2.5, 2.9*	2.0	2.0, 2.1
BMI				
kg/m ²	22.8	22.3, 23.3*	24.7	24.4, 24.9
z-score	0.5	0.4, 0.7	0.7	0.7, 0.8
Overweight (%)	23.5	18.9, 28.8	22.1	20.4, 23.9
Obese (%)	12.1	9.4, 15.4	15.6	13.9, 17.4
Sedentary time				
Total (hours)	6.9	6.8, 6.9	6.4	6.3, 6.5^
Usual bout (minutes)	8.8	8.4, 9.1*	9.1	8.8, 9.3^
MVPA (minutes)	26.1	23.4, 28.8	27.2	25.6, 28.7^
Diet				
Total energy intake (kcal)	2,224	2150, 2298	2,222	2180, 2265
Discretionary snacks (servings)	2.6	2.4, 2.9*	2.3	2.2, 2.4
SSB (servings)	2.0	1.8, 2.1*	2.3	2.2, 2.4
Fruit/Vegetables (servings)	1.8	1.7, 1.9*	1.7	1.6, 1.7
Diet quality score (HEI-2010)	41.8	40.4, 43.2*	40.6	40.1, 41.1

BMI: Body Mass Index; MVPA: Moderate-to-vigorous physical activity; SSB: sugar-sweetened beverages.

^Data available for 1,406

Mean and 95%CI. *Significantly different from those excluded in the analyses (p<0.05)

5.4 Summary

The study presented in Chapter Five was one of the first studies (for any age group) to examine the mediating effects of dietary intake in the relationship between objectively-assessed sedentary time with BMI and metabolic syndrome. Overall, total volume of sedentary time was inversely associated with zBMI (with the association attenuating after adjusting for moderate-to-vigorous physical activity) and no significant associations were observed between usual sedentary bout duration and BMI, and between total volume and usual sedentary bout duration with metabolic syndrome.

Consistent with the candidate's hypothesis, none of the dietary variables were found to mediate the relationship between total volume and bouts of sedentary time with BMI or metabolic syndrome. However, the lack of mediation could simply be due to the null associations observed between the sedentary variables and cardiometabolic outcomes, or due to sedentary time not being matched with dietary intake. Further studies using longitudinal and experimental designs are needed to better understand the role of dietary intake in the sedentary behaviour and cardiometabolic health relationship.

CHAPTER SIX: STUDY THREE

CROSS-SECTIONAL AND PROSPECTIVE MEDIATION
EFFECTS OF DIET ON THE RELATIONSHIP BETWEEN
SEDENTARY BEHAVIOUR AND BODY MASS INDEX
IN ADOLESCENTS

CHAPTER SIX: STUDY THREE

CROSS-SECTIONAL AND PROSPECTIVE MEDIATION EFFECTS OF DIET ON THE RELATIONSHIP BETWEEN SEDENTARY BEHAVIOUR AND BODY MASS INDEX IN ADOLESCENTS

6.1 Overview

Chapters Four and Five examined the cross-sectional mediating effects of dietary intake of the relationship between different types of sedentary behaviour (e.g. TV viewing, total volume of sedentary time and bouts of sedentary time) with BMI and metabolic syndrome in U.S. adolescents. Overall, Chapter Four found only a couple of the dietary variables examined (e.g. sugar-sweetened beverage consumption and fruit and vegetable intake) partially mediated the relationship between TV viewing and metabolic syndrome, whereas Chapter Five found none of the dietary variables mediated the relationship between total volume or bouts of sedentary time with either BMI or metabolic syndrome.

Given the limitations of a cross-sectional design (e.g. unable to detect cause and effect), the temporal order of associations between sedentary behaviour, dietary intake and cardiometabolic health remains unclear, with no study to date exploring this mediating relationship in an adolescent population. Therefore, the following study was undertaken to explore both the cross-sectional and prospective mediating effects of dietary intake on the relationship between sedentary behaviour and BMI in an Australian adolescent cohort.

The following study has been published as a manuscript in *BMC Public Health* (Impact factor: 2.209) and has been formatted according to their requirements. The

citation for the manuscript is: *Fletcher EA, Lamb K, McNaughton SA, Garnett S, Dunstan DW, Baur L, Salmon J. Cross-sectional and prospective mediating effects of dietary intake on the relationship between sedentary behaviour and body mass index in adolescents. BMC Public Health. 2017; 17(1):751.* The manuscript supplementary files can be found directly after the manuscript in *Section 6.3*. The ‘Authorship Statement’ for this manuscript and a copy of the ‘Permission Statement to Publish’ this manuscript are contained in Appendix E.1 and Appendix E.2, respectively.

The author contributions to the manuscript were as follows, with the candidate contributing to all aspects of the manuscript process: Salmon, Baur and Garnett were involved as Chief Investigators of the study; Fletcher, McNaughton, Dunstan and Salmon designed the current study; Fletcher calculated the main variables used in the analyses; Fletcher performed the main analyses with statistical guidance from Lamb; Fletcher wrote the initial draft of the manuscript; and Lamb, McNaughton, Garnett, Dunstan, Baur and Salmon provided critical edits and editions to the manuscript. All authors approved the final manuscript.

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Cross-sectional and prospective mediating effects of dietary intake on the relationship between sedentary behaviour and body mass index in adolescents

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Abstract

Background: Cross-sectional evidence suggests TV viewing, but not objectively-measured sedentary time or bouts of sedentary time, is consistently associated with body mass index (BMI) in adolescents. However, it is unclear whether dietary intake is a potential mediator of these relationships. The aim of this study was to explore the cross-sectional and prospective mediating effects of dietary intake on the association of sedentary behaviour with BMI z-score (zBMI) in a cohort of Australian adolescents.

Methods: Cross-sectional and prospective analyses were conducted in adolescents aged 12–15 years participating in the 2002/03 (baseline) and 2004/05 (follow-up) Nepean Growing Up Study. The independent variables were television (TV) viewing, an objective measure of total sedentary time and average sedentary bout duration, and the outcome variable zBMI. Using the Sobel-Goodman method with bootstrapping, mediation analyses were conducted examining three dietary components (discretionary foods, sugar-sweetened beverages [SSB] and takeaway foods) as mediators of associations between TV viewing and zBMI ($n = 259$) and between total sedentary time and average sedentary bout duration with zBMI ($n = 140$).

Results: No significant cross-sectional or prospective total or direct associations were observed for TV viewing, total sedentary time and average sedentary bout duration with zBMI. However, TV viewing was positively associated with consumption of takeaway foods cross-sectionally ($\beta = 0.06$; 95% CI 0.01 to 0.12), prospectively at baseline ($\beta = 0.07$; 95% CI 0.01 to 0.12) and prospectively at follow-up ($\beta = 0.10$; 95% CI 0.04, 0.16), and average sedentary bout duration was inversely associated with SSB consumption both cross-sectionally ($\beta = -0.36$; 95% CI -0.69 to -0.02) and prospectively at baseline ($\beta = -0.36$; 95% CI -0.70 to -0.02). No mediation effects were identified.

Conclusions: TV viewing, total sedentary time and bouts of sedentary time were not associated cross-sectionally or prospectively with adolescents' zBMI, and three elements of dietary intake (e.g. intake of discretionary foods, SSB and takeaway foods) did not mediate this relationship. The role of dietary intake and sedentary behaviour in relation to adolescent health requires further clarification.

Keywords: Television viewing, Sedentary behaviour, Dietary intake, BMI, Adolescents

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Background

Adolescent obesity is a major public health concern. The combined rates of overweight and obesity among adolescents have increased over the last two decades worldwide [1]. In the United States, the proportion of obese adolescents has risen from 10.5% in 1988–1994 to 20.6% in 2013–2014 [2]. Australia has experienced similar increases with almost one in three adolescents currently overweight or obese [3]. Given that obesity tracks from adolescence to adulthood [4], it is imperative to understand the lifestyle risk factors associated with adolescent obesity, particularly prospectively, in order to inform effective interventions.

Sedentary behaviour – defined as any waking behaviours characterised by low energy expenditure (< 1.5 METS) while in a sitting or reclining posture – has emerged as a new research focus for obesity prevention [5]. High amounts of television (TV) viewing, a common leisure-time sedentary behaviour, during adolescence have both immediate and long-term health consequences, including a higher risk of obesity [6]. However, there are inconsistent associations between total time spent in sedentary time [7, 8], or time spent in periods, or ‘bouts’, of sedentary time [9], and indicators of adiposity (e.g. BMI, waist circumference) in adolescents.

One potential behavioural mechanism that could explain why TV viewing has more consistent associations with body mass index (BMI) when compared to total or bouts of sedentary time in adolescents, is an increase in energy-dense, nutrient poor foods and sugar-sweetened beverages (SSB). For example, TV viewing has consistently been reported to be associated with a higher energy intake, and an increased consumption of discretionary foods, SSB, and fast food/takeaway foods in adolescents [10], whereas few studies have reported associations between objectively-measures of sedentary time with dietary intake [11, 12]. In addition, no study to date has explored whether prolonged bouts of sedentary time are related to dietary intake among adolescents. The latter is important as studies with adults have shown that, independent of how much total sedentary time is accumulated, those with fewer interruptions in sedentary time (i.e., prolonged bouts) have poorer cardiometabolic health profiles [13].

A systematic review examining whether associations between sedentary behaviour and health outcomes in adolescents were independent of dietary intake found TV viewing, screen time and overall sedentary time were positively related to BMI, independent of dietary intake [14]. The systematic review also identified very few studies had specifically examined the mediating role of dietary intake in the TV viewing and BMI relation; with only two studies (out of the 21 studies identified) exploring this and reporting no mediation effects

[15, 16]. However, a major limitation of both of these studies was their cross-sectional design which limits causal inference. Whereas, a longitudinal design would allow both the temporal order of associations to be examined and many other aspects of a mediation model to be explored.

Against this background, the primary aim of the study was to explore both the cross-sectional and prospective mediating effects of the consumption of discretionary foods, SSB and takeaway foods on the association between TV viewing and BMI z-score (zBMI) in a cohort of Australian adolescents, and to examine whether these findings differ when total sedentary time and sedentary bout duration are examined. The secondary aims of the study were to explore the individual associations between the sedentary behaviour variables (TV viewing, sedentary time and sedentary bout duration) and dietary intake variables (discretionary foods, SSB and takeaway foods), as well as their associations with zBMI. Based on the existing evidence, we hypothesised that consumption of discretionary foods, SSB and takeaway foods would partially mediate the cross-sectional and prospective association between TV viewing and zBMI, but would not mediate the cross-sectional or prospective association between total sedentary time and sedentary bouts with zBMI.

Methods

Study design

In 2002/03 (baseline), 348 adolescents aged 12–13 years participated in the Nepean Kids Growing-Up Study. The adolescents were originally part of a birth cohort study (the “Nepean Study”) which involved 2314 infants born between 1989 and 1990 at the Nepean Hospital (western Sydney, Australia). Written consent was obtained from the parent or guardian and assent from the adolescent. The Ethics Committees of The Children’s Hospital at Westmead and Wentworth Area Health Service gave ethical approval. Full details about the original study and eligibility criteria have been previously published [17]. Briefly, the study involved adolescents attending the clinic at Nepean Hospital, where they had their height and weight measured and completed a questionnaire on their demographics and physical activity levels, and a semi-quantitative food frequency questionnaire (FFQ). Afterwards, adolescents wore an accelerometer for 7 days during all waking hours. In 2004/05, the adolescents were recontacted and invited to participate in the follow-up study. In total, 63 adolescents were unable to be contacted or withdrew from the study, leaving 285 adolescents participating at both time points.

Outcome variable (zBMI)

Height and weight were measured at both time points by a trained research assistant and study dietitian at the

clinic. Height was measured to the nearest 0.1 cm and weight was measured without shoes and in light clothing to the nearest 0.1 kg with electronic scales (Wedderburn, Summer Hill, NSW, Australia). Height and weight were used to calculate each participants' BMI and zBMI was determined using the age- and sex-specific CDC 2000 reference data [18]. Overweight and obesity was determined using the International Obesity Task Force (IOTF) criteria [19]. In all analyses, zBMI was treated as a continuous variable.

Independent variables (sedentary behaviour)

Adolescents completed a self-report questionnaire on their time spent watching TV (hours/day) on a usual school day (Monday to Friday) and a usual weekend day (Saturday and Sunday). The questionnaire has previously been shown to have good to excellent reliability (percentage agreement = 70%–99%) [20]. To calculate average daily hours spent watching TV over a usual week, daily weekday TV hours was multiplied by five and daily weekend TV hours was multiplied by 2, then summed together and divided by seven.

Sedentary time was measured objectively by an ActiGraph AM-7164 accelerometer (ActiGraph Inc, Florida). At both time points, adolescents were asked to wear the monitor on their right hip during all waking hours for 7 days, except when bathing, swimming and sleeping. Data were downloaded in 1-min epochs and non-wear time was defined as at least 20 min of zero counts. Sedentary time was defined as all wear-time minutes with an average activity count of ≤ 100 counts per minute (cpm), and was standardised for wear time using the residual method [21]. Average sedentary bout duration was calculated by summing all uninterrupted minutes ≤ 100 cpm, and then taking the midpoint of all sedentary bouts that lie on the accumulation curve for each individual [22]. Analyses were limited to participants who had ≥ 8 h of wear time on ≥ 3 week days and ≥ 7 h of wear time on ≥ 1 weekend day [23].

Mediating variables (dietary intake)

Usual dietary intake was measured using a 56-item semi-quantitative FFQ, which was developed based on data from the 1995 Australian National Nutrition Survey [24]. Adolescents were asked to report how often they ate certain foods and beverages (either in times per week or per day) over the previous 7 days. The 8-item frequency response scale was converted to times per week as follows: 1) "not consumed last week" = 0; 2) "consumed once last week" = 0.143; 3) "consumed 2–3 times last week" (average number used) = 0.357; 4) "consumed 4–6 times last week" (average number used) = 0.714; 5) "consumed once a day" = 1; 6) "consumed 2 times a day" = 2; 7) "consumed 3 times a

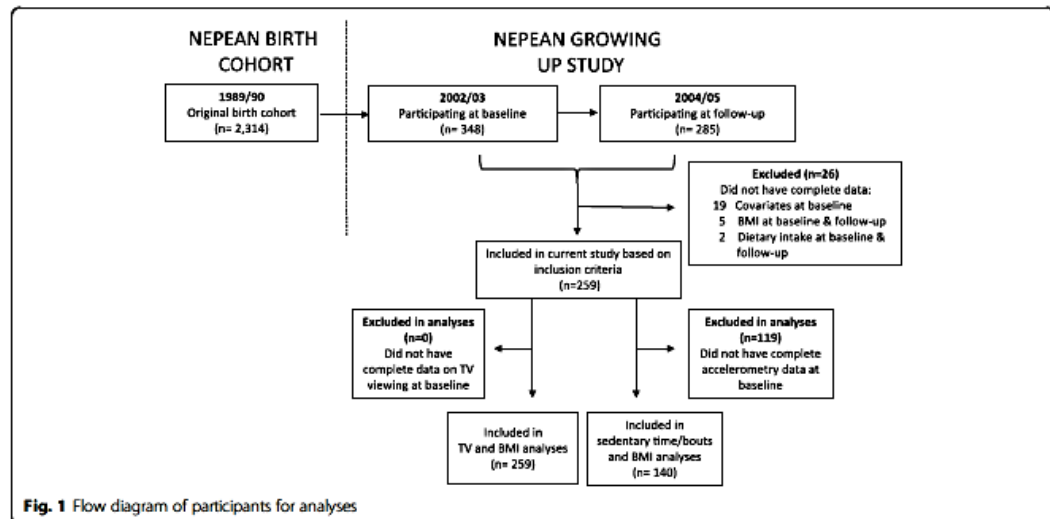
day" = 3; 8) "consumed 4–6 times a day" (average number used) = 5. For the analyses, a combination of food and beverage items were summed together to create three dietary mediators at both time points: 1) frequency of consuming discretionary foods, which included any savoury or plain biscuits, sweet pastries, cakes, doughnuts, chocolate, confectionary, and potato chips; 2) frequency of consuming SSB, which included non-diet soft drink, non-diet cordial and fruit juice and not sweetened milk drinks or energy drinks; and 3) frequency of consuming takeaway foods, which included savoury pastries (e.g. meat pies and sausage rolls), hamburgers, pizza, hot chips and spring rolls/dim sims. The FFQ was tested for reproducibility and overall showed fair to excellent reliability for sweet snacks (ICC = 0.61), savoury snacks (ICC = 0.63), SSBs (ICC = 0.77), and fast food (ICC = 0.44). Frequency of consuming discretionary foods and frequency of consuming SSB were multiplied by 7 to convert to times per day. Participants with missing data for any of the dietary items listed were excluded ($n = 2$). In all analyses, the dietary mediators were treated as continuous variables.

Covariates

The covariates considered for the analyses included age at baseline, sex, maternal education (an indicator for family socioeconomic status), pubertal status and accelerometry-measured moderate-to-vigorous physical activity (MVPA) collected at baseline. Maternal education was collapsed into three categories: "low" (some secondary education or less); "medium" (completing secondary school, an apprenticeship or technical certificate); and "high" (university or tertiary qualification). Pubertal status was self-assessed using the 'Tanner Stages' of breast development and commencement of menses (girls) and pubic hair and genitalia (boys) [25]. For analyses, participants were categorised as early puberty, mid-pubertal, late-pubertal and post-pubertal. Accelerometry-measured MVPA at baseline was calculated based on the Freedson accelerometer age-cut points [26] and considered as a covariate for the analyses involving sedentary time and sedentary bouts with zBMI.

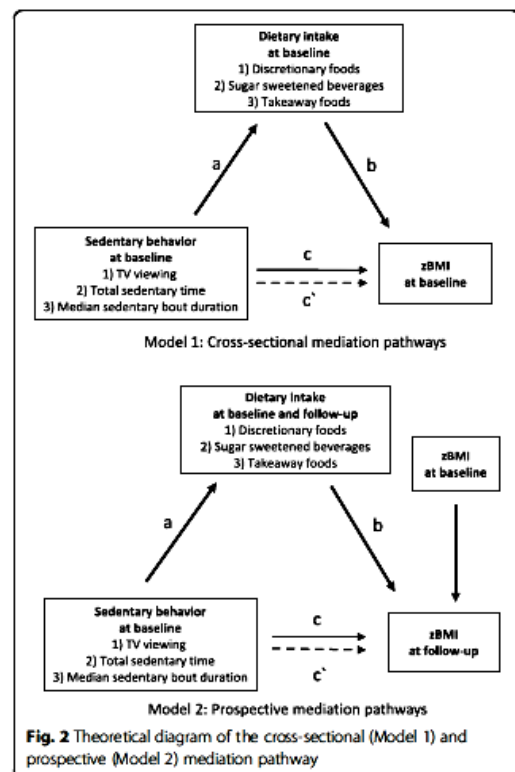
Statistical analyses

As shown in Fig. 1, to be included in the analyses involving TV viewing and zBMI, participants were required to have complete data for age, sex, maternal education, pubertal status, dietary intake and TV viewing at baseline, and complete data for zBMI at baseline and follow-up ($n = 259$). To examine the association of total and bouts of sedentary time, dietary intake and zBMI, separate analyses were undertaken from a subsample of participants who met the previous inclusion criteria as well as



meeting the accelerometry inclusion criteria at baseline ($n = 140$). Prior to the main analyses, all variables were checked for normality. Discretionary foods, SSB and takeaway food intake at baseline were not normally distributed and were log-transformed.

Analyses were conducted using Stata/SE v14.0 (StataCorp LP, College Station, Texas, 2015). Figure 2 illustrates two theoretical models of the cross-sectional and prospective mediation pathways examined [27]. For all mediation analyses, the Sobel-Goodman mediation method with bootstrapping with 5000 replications was used to estimate standard errors and 95% confidence intervals [28]. For the cross-sectional analyses, only baseline variables were used to test the following associations: 1) association between the independent variable and the mediator (a-coefficient pathway); 2) association between the mediator and the outcome variable, adjusting for the independent variable (b-coefficient pathway); 3) total association between the independent variable and outcome variable (c-coefficient pathway); 4) direct association between the independent variable and outcome variable, accounting for each mediator (c'-coefficient pathway); and 5) indirect association (e.g. mediating effect) of the mediator on the independent variable and outcome variable. For the prospective analyses, similar pathways were tested. However, dietary intake at baseline and dietary intake at follow-up were examined separately as potential mediators in the associations between the independent variable at baseline and the outcome variable at follow-up. All analyses were adjusted for age, sex, pubertal status and maternal education, with objectively-measured MVPA adjusted for in the analyses involving total and bouts of sedentary time



with zBMI. The prospective analyses were additionally adjusted for zBMI at baseline. The significance level was set at $p < 0.05$ for all statistical tests.

Results

Overall, 259 and 140 adolescents were included in the TV viewing and zBMI, and the sedentary time and zBMI analytic samples, respectively (Table 1). Those excluded in the TV viewing and zBMI analyses had mothers with a lower maternal education and those excluded in the

sedentary time and zBMI analyses were older, had a lower maternal education and had a higher proportion of overweight participants (Additional file 1: Tables S1 and S2 in the online supplement file).

Table 1 Baseline characteristics of participants included in analyses

Variables	TV and zBMI (n = 259)	Sedentary time and zBMI (n = 140)
Age, years	12.9 (12.9, 13.0)	12.9 (12.9, 13.0)
Sex, %		
Male	47.5 (41.4, 53.6)	50.0 (41.7, 58.3)
Female	52.5 (46.4, 58.4)	50.0 (41.7, 58.3)
Maternal education, %		
Low	6.9 (4.4, 10.8)	5.0 (2.3, 10.2)
Medium	65.3 (59.2, 70.8)	62.8 (54.4, 70.5)
High	27.8 (22.7, 33.6)	32.1 (24.8, 40.4)
Pubertal status, %		
Early puberty	19.3 (8.0, 4.6)	14.3 (35.1, 56.2)
Mid-pubertal	35.5 (30.0, 41.6)	41.4 (33.5, 50.0)
Late-pubertal	35.5 (30.0, 41.6)	35.0 (27.5, 43.4)
Post-pubertal	27.0 (21.9, 32.8)	22.1 (16.0, 30.0)
BMI		
kg/m ²	20.7 (20.2, 21.3)	20.3 (19.7, 21.0)
z-score	0.4 (0.3, 0.5)	0.3 (0.1, 0.5)
Overweight, %	18.9 (14.6, 24.2)	13.6 (8.8, 20.4)
Obese, %	9.2 (6.3, 13.5)	9.2 (5.4, 15.4)
Sedentary behaviour		
TV viewing (hours/day)	3.2 (3.0, 3.4)	
Total sedentary time (hours/day)		6.1 (6.0, 6.2)
Average sedentary bout (minutes/day)		6.6 (6.5, 6.6)
MVPA (minutes/day)		48.2 (35.0, 61.4)
Dietary intake		
Discretionary foods (freq/day)	2.4 (2.2, 2.6)	2.4 (2.1, 2.7)
Sugar-sweetened beverages (freq/day)	2.1 (1.8, 2.3)	1.9 (1.6, 2.2)
Takeaway food (freq/week)	3.4 (3.1, 3.6)	3.2 (2.8, 3.6)

BMI body mass index, zBMI BMI z-score, TV television, MVPA moderate-to-vigorous physical activity; freq: frequency
Footnote: Maternal education: low = some secondary education or less; medium = completing secondary school, an apprenticeship or technical certificate; high = university or tertiary qualification. Pubertal status self-assessed using the 'Tanner Stages' [25]. BMI z-score determined using the age- and sex-specific CDC 2000 reference data [18]. Overweight and obesity determined using the International Obesity Task Force (IOTF) criteria [19]

Associations between TV viewing, dietary intake and zBMI

Cross-sectionally

There was no evidence of total or direct cross-sectional associations of TV viewing with zBMI (Table 2). A small, positive association was observed between hours spent watching TV per day and frequency of consuming take-away foods each week (a-coefficient pathway); with each additional hour of TV viewing, adolescents consumed an additional 0.06 serves (95% CI 0.01 to 0.12; $p < 0.05$) of takeaway foods each week. An inverse association was also observed for consuming discretionary foods each day and zBMI (b-coefficient pathway); with each additional serving of discretionary foods consumed each day, zBMI was lower by -0.39 units (95% CI -0.65 to -0.13 ; $p < 0.01$). None of the dietary variables were significant mediators of the cross-sectional association of TV viewing with zBMI.

Prospectively

There was no evidence of total or direct prospective associations between TV viewing at baseline and zBMI at follow-up. Similar to the cross-sectional associations, a positive prospective association was observed between hours spent watching TV per day and frequency of consuming takeaway foods each week at baseline and at follow-up. For example, for each additional hour of TV viewing, adolescents consumed an additional 0.07 (95% CI 0.01 to 0.12; $p < 0.05$) serves of take-away foods each week at baseline, and an additional 0.10 (95% CI 0.04 to 0.16; $p < 0.05$) serves of take-away foods each week at follow-up. However, no significant association remained for any of the dietary variables consumed at baseline and at follow-up with zBMI at follow-up (b-coefficient pathway). None of the dietary variables were significant mediators of the prospective association between TV viewing and zBMI.

Associations between total sedentary time, dietary intake and zBMI

Cross-sectionally

There was no evidence of total or direct cross-sectional associations for total sedentary time (hours/day) and zBMI, or between total sedentary time and any of the dietary mediators (a-coefficient pathway) (Table 3). An inverse association was observed between frequency of consuming discretionary foods per day and zBMI; with each additional discretionary food consumed each week, zBMI was lower by -0.42 units (95% CI -0.77 to -0.07 ; $p < 0.05$) (b-coefficient). There were no significant mediating effects for any of the dietary variables in the cross-sectional association of total sedentary time with zBMI.

Table 2 Cross-sectional^a and prospective^b associations of dietary intake (mediator), TV viewing (independent) and zBMI (outcome) (n = 259)

Outcome: zBMI Independent: TV viewing	c'-coefficient (direct) β (95% CI)	a-coefficient β (95% CI)	b coefficient β (95% CI)	ab/indirect (mediated/indirect) ^f	
				Uncorrected β (95% CI)	Bias corrected β (95% CI)
Cross-sectional mediators					
Discretionary food (freq/day)	0.05 (-0.02, 0.13)	0.01 (-0.03, 0.05)	-0.39 (-0.65, -0.13)**	-0.04 (-0.18, 0.10)	-0.04 (-0.23, 0.12)
SSB (freq/day)	0.05 (-0.03, 0.13)	0.02 (-0.03, 0.07)	0.03 (-0.15, 0.22)	0.01 (-0.04, 0.05)	0.01 (-0.07, 0.09)
Takeaway food (freq/week)	0.05 (-0.02, 0.13)	0.06 (0.01, 0.12)*	-0.07 (-0.24, 0.09)	-0.05 (-0.16, 0.07)	-0.05 (-0.22, 0.06)
Prospective mediators					
At baseline					
Discretionary foods (freq/day)	-0.01 (-0.04, 0.02)	0.01 (-0.02, 0.05)	0.02 (-0.09, 0.13)	0.01 (-0.01, 0.02)	0.01 (-0.02, 0.04)
SSB (freq/day)	-0.01 (-0.04, 0.02)	0.02 (-0.03, 0.07)	0.03 (-0.04, 0.11)	0.01 (-0.02, 0.03)	0.01 (-0.02, 0.04)
Takeaway food (freq/week)	-0.01 (-0.04, 0.02)	0.07 (0.01, 0.12)*	-0.01 (-0.07, 0.06)	-0.01 (-0.04, 0.05)	-0.01 (-0.06, 0.05)
At follow-up					
Discretionary foods (freq/day)	-0.01 (-0.04, 0.02)	0.02 (-0.02, 0.07)	-0.04 (-0.13, 0.06)	-0.01 (-0.04, 0.02)	-0.01 (-0.06, 0.02)
SSB (freq/day)	-0.01 (-0.04, 0.02)	-0.01 (-0.05, 0.04)	0.03 (-0.06, 0.11)	-0.001 (-0.01, 0.01)	-0.001 (-0.03, 0.02)
Takeaway food (freq/week)	-0.01 (-0.04, 0.02)	0.10 (0.04, 0.16)*	0.02 (-0.05, 0.08)	0.02 (-0.05, 0.08)	0.02 (-0.04, 0.09)

^aCross-sectional total effects (c-pathway) of TV viewing and zBMI $\beta = 0.05$ (95% CI -0.03 to 0.13), adjusting for age, sex, mother's education and pubertal status.

^bProspective total effects (c-pathway) of TV viewing and zBMI $\beta = -0.01$ (95% CI -0.04 to 0.02), adjusting for age, sex, mother's education, pubertal status and zBMI at baseline.

^cDue to the small units of measure, the indirect effects have been multiplied by 10

Significant ** $p < 0.01$, * $p < 0.05$. TV: television; SSB: sugar-sweetened beverages; zBMI: body mass index z-score; freq: frequency

Prospectively

Total sedentary time at baseline was not significantly associated with zBMI at follow-up, accounting for mediation by dietary intake at baseline and at follow-up. No significant associations were also observed for any of the total, direct and indirect pathways, nor with the a-coefficient and b-coefficient pathways.

Associations between average sedentary bout duration, dietary intake and zBMI

Cross-sectionally

No significant total or direct cross-sectional associations were observed for average sedentary bout duration (minutes/day) with zBMI (Table 4). Average sedentary bout duration was inversely related to frequency of

Table 3 Cross-sectional^a and prospective^b associations of dietary intake (mediator), sedentary time (independent) and zBMI (outcome) (n = 140)

Outcome: zBMI Independent: total sedentary time	c'-coefficient (direct) β (95% CI)	a-coefficient β (95% CI)	b coefficient β (95% CI)	ab/indirect (mediated/indirect) ^f	
				Uncorrected β (95% CI)	Bias corrected β (95% CI)
Cross-sectional mediators					
Discretionary food (freq/day)	0.27 (-0.03, 0.58)	-0.01 (-0.15, 0.14)	-0.42 (-0.77, -0.07)*	0.01 (-0.61, 0.62)	0.01 (-0.62, 0.69)
SSB (freq/day)	0.27 (-0.05, 0.58)	-0.16 (-0.38, 0.06)	-0.04 (-0.29, 0.20)	0.07 (-0.33, 0.46)	0.07 (-0.31, 0.61)
Takeaway food (freq/week)	0.27 (-0.05, 0.58)	-0.06 (-0.30, 0.18)	-0.04 (-0.26, 0.18)	0.02 (-0.13, 0.18)	0.02 (-0.36, 0.35)
Prospective mediators					
At baseline					
Discretionary food (freq/day)	0.05 (-0.07, 0.17)	0.03 (-0.12, 0.17)	0.03 (-0.10, 0.17)	0.01 (-0.05, 0.07)	0.01 (-0.08, 0.15)
SSB (freq/day)	0.05 (-0.06, 0.17)	-0.15 (-0.37, 0.07)	0.02 (-0.07, 0.11)	-0.03 (-0.17, 0.11)	-0.03 (-0.22, 0.13)
Takeaway food (freq/week)	0.05 (-0.07, 0.17)	-0.05 (-0.30, 0.19)	-0.03 (-0.12, 0.05)	0.02 (-0.08, 0.11)	0.02 (-0.12, 0.16)
At follow-up					
Discretionary food (freq/day)	0.05 (-0.07, 0.17)	0.08 (-0.09, 0.24)	0.01 (-0.11, 0.13)	0.01 (-0.05, 0.10)	0.01 (-0.16, 0.17)
SSB (freq/day)	0.05 (-0.06, 0.17)	-0.04 (-0.23, 0.15)	0.01 (-0.10, 0.12)	-0.01 (-0.05, 0.04)	-0.01 (-0.11, 0.09)
Takeaway food (freq/week)	0.05 (-0.06, 0.17)	0.21 (-0.57, 0.47)	-0.01 (-0.08, 0.07)	-0.01 (-0.16, 0.15)	-0.01 (-0.19, 0.17)

^aCross-sectional total effects (c-pathway) of total sedentary time and zBMI $\beta = 0.27$ (95% CI -0.04 to 0.58), adjusting for age, sex, mother's education and pubertal status.

^bProspective total effects (c-pathway) of total sedentary time and zBMI $\beta = 0.05$ (95% CI -0.06 to 0.17), adjusting for age, sex, mother's education, pubertal status and zBMI at baseline.

^cDue to the small units of measure, the indirect effects have been multiplied by 10. Significant * $p < 0.05$. SSB: sugar-sweetened beverages; zBMI: body mass index z-score; freq: frequency

Table 4 Cross-sectional^a and prospective^b associations of dietary intake (mediator), sedentary bouts (independent) and zBMI (outcome) (*n* = 140)

Outcome: zBMI	c'-coefficient (direct) β (95% CI)	a-coefficient β (95% CI)	b-coefficient β (95% CI)	ab/indirect (mediated/indirect) ^c	
Independent: sedentary bouts				Uncorrected β (95% CI)	Bias-corrected β (SE)
Cross-sectional mediators					
Discretionary food (freq/day)	0.12 (-0.36, 0.61)	-0.08 (-0.31, 0.15)	-0.42 (-0.77, -0.06)*	0.32 (-0.67, 1.32)	0.32 (-0.55, 1.37)
SSB (freq/day)	0.14 (-0.36, 0.64)	-0.36 (-0.69, -0.02)*	-0.06 (-0.30, 0.19)	0.20 (-0.71, 1.11)	0.20 (-0.68, 0.12)
Takeaway food (freq/week)	0.16 (-0.33, 0.65)	0.03 (-0.34, 0.41)	-0.05 (-0.28, 0.18)	-0.02 (-0.21, 0.18)	-0.02 (-0.62, 0.34)
Prospective mediators					
At baseline					
Discretionary food (freq/day)	0.08 (-0.10, 0.26)	-0.06 (-0.29, 0.17)	0.04 (-0.10, 0.17)	-0.02 (-0.14, 0.10)	-0.02 (-0.24, 0.14)
SSB (freq/day)	0.09 (-0.09, 0.27)	-0.36 (-0.70, -0.02)*	0.02 (-0.07, 0.11)	-0.09 (-0.42, 0.24)	-0.09 (-0.52, 0.24)
Takeaway food (freq/week)	0.08 (-0.10, 0.26)	0.04 (-0.34, 0.41)	-0.04 (-0.12, 0.04)	-0.01 (-0.15, 0.13)	-0.01 (-0.27, 0.16)
At follow-up					
Discretionary food (freq/day)	0.08 (-0.10, 0.26)	-0.08 (-0.34, 0.17)	0.02 (-0.10, 0.14)	-0.02 (-0.12, 0.09)	-0.02 (-0.28, 0.16)
SSB (freq/day)	0.08 (-0.10, 0.26)	0.11 (-0.18, 0.40)	0.01 (-0.010, 0.11)	0.01 (-0.11, 0.12)	0.01 (-0.19, 0.20)
Takeaway food (freq/week)	0.08 (-0.10, 0.26)	0.06 (-0.35, 0.48)	-0.01 (-0.08, 0.07)	-0.01 (-0.05, 0.04)	-0.01 (0.18, 0.17)

^aCross-sectional total effects (c-pathway) of average sedentary bout and zBMI β = 0.16 (95% CI -0.33 to 0.65), adjusting for age, sex, mother's education and pubertal status. ^bProspective total effects (c-pathway) of average sedentary bout and zBMI β = 0.08 (95% CI -0.10 to 0.26), adjusting for age, sex, mother's education, pubertal status and zBMI at baseline. ^cDue to the small units of measure, the indirect effects have been multiplied by 10. Significant **p* < 0.05. SSB: sugar-sweetened beverages; zBMI: body mass index z-score; freq: frequency

consuming SSB each day (a-coefficient pathway); with each additional minute spent in a sedentary bout, the frequency of consuming SSB was lower by nearly half a serve each day (β = -0.36, 95% CI -0.69 to -0.02; *p* < 0.05). An inverse association was also observed for frequency of consuming discretionary foods each week and zBMI (b-coefficient pathway); with each additional discretionary food consumed each week, zBMI was lower by -0.42 units (95% CI -0.77 to -0.06; *p* < 0.05). None of the dietary variables significantly mediated cross-sectional associations of sedentary bout duration with zBMI.

Prospectively

When examining the prospective associations between average sedentary bout duration at baseline and zBMI at follow-up, no significant total or direct associations were observed. However, a significant inverse association remained for average sedentary bout duration with frequency of consuming SSB at baseline (β = -0.36; 95% CI -0.70 to -0.02; *p* < 0.05), but not at follow-up for the a-coefficient pathways. No significant associations were observed for the b-coefficient pathways or mediating effects.

Discussion

This study found no evidence of direct or indirect associations for TV viewing, total sedentary time and average sedentary bout duration with adolescents' zBMI, either cross-sectionally or prospectively. Although some of the dietary variables were independently associated

with TV viewing, average sedentary bout duration and zBMI, none of the dietary variables significantly mediated the relationships between the sedentary variables and zBMI cross-sectionally or prospectively.

The null finding for the association of TV viewing with zBMI in the current study is in contrast to previous studies in youth that have consistently shown significant and positive associations both cross-sectionally [6] and prospectively [29, 30]. The differences in findings could be attributed to the homogeneity of the current sample being examined, with higher than average number of hours spent watching TV [31] and a lower zBMI compared to the population average [32]. The null association for total sedentary time and average sedentary bout duration with zBMI is supported by some previous studies [8, 33–35], but not others [7]. Research examining accelerometer-measured sedentary time with health indicators among children and youth appears to be mixed; it is unclear whether an association exists in only some populations or if there are inconsistencies in measuring sedentary time and/or the analytical approaches undertaken.

The positive association observed for TV viewing with the consumption of takeaway foods, both cross-sectionally and prospectively is consistent with previous research [36, 37]. This link could be partially explained by the large extent of TV advertising of foods high in fat and energy during peak times when children and adolescents are likely to be watching TV [38]. In contrast, no evidence of an association was observed for total sedentary time with any of the dietary

variables. The null finding is consistent with previous literature in youth [11, 12] where, compared to TV viewing, fewer significant associations are observed for total sedentary time with elements of a less healthy diet. The null finding could be due to the measure used to capture total sedentary time. For example, accelerometers measure *all* time spent being sedentary, and thus may capture times where adolescents may not be eating/drinking (e.g. sitting in school, sitting in the car). In addition, due to accelerometers being unable to determine posture (e.g. standing still versus sitting), time spent standing may have been included in total sedentary time and thus may diminish the opportunity to engage in an eating occasion. Unexpectedly, the study found higher sedentary bout duration was inversely associated with a lower consumption of SSB, both cross-sectionally and prospectively. One possible reason for this could be due to the adolescents not breaking up their sedentary time in order to retrieve a SSB from another room (e.g. the kitchen, school canteen). However, given the current study is one of the first to examine individual associations between sedentary bout duration and dietary intake, further research is urgently needed in this area.

In contrast, the cross-sectional, inverse association found for discretionary foods with zBMI, and the null finding for SSB and takeaway foods is in contrast with previous studies that have found positive associations for unhealthy dietary intake with BMI [39, 40]. Our findings could be a consequence of under-reporting, where overweight or obese children and adolescents have been found to under-report their energy intake by 20–40% [41]. Alternatively, it is possible that some participants in the current study with a higher zBMI may have changed their behaviour by decreasing their discretionary food intake over time as a strategy to manage their weight.

Only one other study has explored the prospective associations between TV viewing, dietary intake and BMI; however, that study examined a younger population of pre-school children aged 0–5 years over a 2-year period [42]. In contrast to the current study, Fuller-Tyszkiewicz et al. reported a significant positive association between TV viewing and BMI that was bi-directional, with those children characterised with high amounts of TV viewing having higher BMI, and children with higher BMI watching a greater amount of TV. In addition, that study reported the prospective associations between TV viewing and BMI among 4 year olds were mediated by discretionary foods and soft drink consumption [42]. The differences in findings between the current study and previous study could be contributed to the different study populations and dietary mediators being examined, and that the previous study only examined TV viewing and BMI. Thus, further prospective studies are needed to explore whether dietary intake mediates the relationship between various sedentary behaviours

(both subjective and objectively measured) and health indicators (e.g. BMI, metabolic syndrome).

To our knowledge, the current study is the first to examine both the cross-sectional and prospective mediating effects of dietary intake on the association between sedentary behaviour and zBMI in an adolescent population, and the first to examine this using objective measures of sedentary time. Other strengths include adjustment for a variety of confounders, including maternal education and adolescent pubertal status, and examining the dietary mediators (discretionary foods, SSB, takeaway foods) separately in all models. Limitations of the study include participants self-reporting their dietary intake using an FFQ and hours spent watching TV, and the low sample size for mediation analyses. In addition, the semi-quantitative FFQ used in the current study was limited with the number of healthy food items included (e.g. fruit and vegetables) and did not have information on portion sizes. There were also differences in those that were included and excluded in the analysis of this study and thus may limit the representativeness of the findings. Further, the data presented in the current study was collected more than a decade ago and thus the behaviours reported in the study may not reflect the contemporary sedentary and dietary behaviours adolescents are engaging in today.

Conclusion

In conclusion, despite identifying some significant associations between TV viewing and average sedentary bout duration with frequency of consuming takeaway foods and SSB, and between frequency of consuming discretionary foods and zBMI, no significant associations were observed for any of the sedentary behaviour variables with zBMI, either cross-sectionally or prospectively. In addition, none of the dietary variables were found to be significant mediators of the associations between sedentary behaviour and zBMI. Given the unacceptably high levels of adolescent overweight and obesity, further studies are warranted to elucidate the complex relationships between TV viewing, sedentary time, dietary intake and health indicators.

Additional file

Additional file 1: Supplement file. Comparison of baseline characteristics of participants included in the TV viewing and zBMI analyses compared to those excluded in the analyses. (PDF 29 kb)

Abbreviations

BMI: Body mass index; Cpm: Counts per minute; FFQ: Food frequency questionnaire; MVPA: Moderate-to-vigorous physical activity; SSB: Sugar-sweetened beverages; TV: Television; zBMI: Body mass index z-score

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Availability of data and materials

The dataset supporting the conclusions of this article has not been approved to be made publicly available by the ethics committees. To require information about the content and analyses of the dataset, contact Sarah Garnett: sarah.garnett@health.nsw.gov.au

Authors' contributions

JS, LB, and SG were involved in the conception and design of the study. EF carried out the data analysis with statistical guidance from KL. EF led the writing of the article, with contributions from authors KL, SM, JS, LB, SG, and DD. All authors read and approved the final manuscript.

Ethics approval and consent to participate

Ethical approval was provided by the Ethics Committees of The Children's Hospital at Westmead (Project No. 2001/095) and Wentworth Area Health Service (Registration No. 2001/065). Written consent was obtained from the adolescent's parent or guardian and each adolescent signed a study agreement form.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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6.3 Manuscript supplementary files

The supplementary file supporting the manuscript “*Cross-sectional and prospective mediating effects of dietary intake on the relationship between sedentary behaviour and body mass index in adolescents*” can be found on the following pages.

The supplementary files include:

- Table S1: Comparison of baseline characteristics of participants included in the TV viewing and zBMI analyses compared to those excluded in the analyses
- Table S2: Comparison of baseline characteristics of participants included in the sedentary time and zBMI analyses compared to those excluded in the analyses

Table S1: Comparison of mean (95%CI) baseline characteristics of participants included in the TV viewing and zBMI analyses (n=259) compared to those excluded in the analyses (n=88)

Variables	Included (n= 259)	Excluded (n= 88)
Age, years	12.9 (12.9, 13.0)	13.0 (12.9, 13.0)
Sex, %		
Male	47.5 (41.4, 53.6)	56.2 (45.6, 66.3)
Female	52.5 (46.4, 58.4)	43.8 (33.7, 54.5)
Maternal education, %		
Low	6.9 (4.4, 10.8)	18.6 (11.6, 28.5)*
Medium	65.3 (59.2, 70.8)	64.0 (53.1, 73.5)
High	27.8 (22.7, 33.6)	17.4 (10.7, 27.2)
Pubertal status, %		
Early puberty	19.3 (8.0, 4.6)	0.0 (0.0, 0.0)
Mid-pubertal	35.5 (30.0, 41.6)	31.4 (21.5, 43.5)
Late-pubertal	35.5 (30.0, 41.6)	44.3 (32.9, 56.3)
Post-pubertal	27.0 (21.9, 32.8)	24.3 (15.5, 36.0)
BMI		
kg/m ²	20.7 (20.2, 21.3)	20.8 (19.9, 21.7)
z-score	0.4 (0.3, 0.5)	0.4 (0.2, 0.6)
Overweight, %	18.9 (14.6, 24.2)	23.9 (56.4, 76.2)
Obese, %	9.2 (6.3, 13.5)	9.1 (4.5, 17.3)
Sedentary behavior		
TV viewing (hours/day)	3.2 (3.0, 3.4)	3.0 (2.7, 3.3)
Dietary intake		
Discretionary foods (freq/day)	2.4 (2.2, 2.6)	2.3 (2.0, 2.7)
Sugar-sweetened beverages (freq/day)	2.1 (1.8, 2.3)	2.0 (14.6, 2.4)
Takeaway food (freq/week)	3.4 (3.1, 3.6)	3.5 (2.9, 4.1)

TV: television; MVPA: moderate-to-vigorous physical activity; freq: frequency

*Significant $p < 0.05$

Table S2: Comparison of mean (95%CI) baseline characteristics of participants

included in the sedentary time and zBMI analyses (n=140) compared to those excluded in the analyses (n=88)

Variables	Included (n= 140)	Excluded (n= 207)
Age, years	12.9 (12.9, 13.0)	13.0 (12.9, 13.0)*
Sex, %		
Male	50.0 (41.7, 58.3)	49.5 (42.7, 56.3)
Female	50.0 (41.7, 58.3)	50.5 (43.7, 57.3)
Maternal education, %		
Low	5.0 (2.3, 10.2)	13.2 (9.2, 18.6)*
Medium	62.8 (54.4, 70.5)	66.3 (59.5, 72.5)
High	32.1 (24.8, 40.4)	20.5 (15.5, 26.6)
Pubertal status, %		
Early puberty	14.3 (35.1, 56.2)	1.6 (0.5, 4.8)
Mid-pubertal	41.4 (33.5, 50.0)	29.6 (23.5, 36.6)
Late-pubertal	35.0 (27.5, 43.4)	39.2 (32.4, 46.4)
Post-pubertal	22.1 (16.0, 30.0)	29.6 (23.5, 36.6)
BMI		
kg/m ²	20.3 (19.7, 21.0)	21.0 (20.4, 21.6)
z-score	0.3 (0.1, 0.5)	0.5 (0.3, 0.6)
Overweight, %	13.6 (8.8, 20.4)	24.6 (19.2, 31.0)*
Obese, %	9.2 (5.4, 15.4)	9.2 (5.9, 14.0)
Sedentary behavior		
Total sedentary time (hours/day)	6.1 (6.0, 6.2)	5.8 (5.3, 6.2)
Average sedentary bouts (mins/day)	6.6 (6.5, 6.6)	6.6 (6.5, 6.6)
MVPA (mins/day)	48.2 (35.0, 61.4)	40.8 (34.2, 47.4)
Dietary intake		
Discretionary foods (freq/day)	2.4 (2.1, 2.7)	2.3 (2.1, 2.5)
Sugar-sweetened beverages (freq/day)	1.9 (1.6, 2.2)	2.2 (1.9, 2.4)
Takeaway food (freq/week)	3.2 (2.8, 3.6)	3.5 (3.1, 3.9)

TV: television; MVPA: moderate-to-vigorous physical activity; freq: frequency

*Significant $p < 0.05$

6.4 Summary

The study presented in Chapter Six is the first study to examine both the cross-sectional and prospective mediating effects of dietary intake on the association between various sedentary behaviours (e.g. TV viewing, total volume and bouts of sedentary time) and BMI in a cohort of Australian adolescents. Overall, no significant cross-sectional or prospective associations were observed for any of the sedentary behaviour variables and BMI.

In contrast to the candidate's hypothesis, none of the dietary variables (e.g. frequency of consumption of discretionary foods, sugar-sweetened beverages and takeaway foods) mediated the relationship between TV viewing and BMI, either cross-sectionally and prospectively, nor did any of the dietary variables mediate the relationship when total volume and bouts of sedentary time were examined. These findings, along with the findings presented in Chapter Four and Five, highlight the complex relationships observed between TV viewing, sedentary time, dietary intake and cardiometabolic health indicators. Further research using experimental designs may allow a better understanding of the role dietary intake may play in the sedentary behaviour and cardiometabolic health relationship.

CHAPTER SEVEN: STUDY FOUR

EFFECTS OF BREAKING UP SITTING ON
ADOLESCENTS' POSTPRANDIAL GLUCOSE AFTER
CONSUMING MEALS VARYING IN ENERGY:
AN EXPERIMENTAL TRIAL

CHAPTER SEVEN: STUDY FOUR

EFFECTS OF BREAKING UP SITTING ON ADOLESCENTS' POSTPRANDIAL GLUCOSE AFTER CONSUMING MEALS VARYING IN ENERGY: AN EXPERIMENTAL TRIAL

7.1 Overview

Chapters Four, Five and Six have examined the cross-sectional and prospective mediating effects of dietary intake of the relationship between different types of sedentary behaviours (e.g. TV viewing, total sedentary time and bouts of sedentary time) with BMI and metabolic syndrome in U.S. and Australian adolescent cohorts. These observational findings provided mixed findings into the mediating effects of dietary intake in the sedentary behaviour and cardiometabolic health relationship. For example, while Chapter Four found some of the dietary variables to partially mediate the relationship between TV viewing and metabolic syndrome, Chapters Five and Six found no mediating effects with any of the dietary variables in the relationship between TV viewing, total volume and bouts of sedentary time with either BMI or metabolic syndrome.

Due to the observational design of the studies presented in Chapters Four to Six, it remains unclear how dietary intake interacts with sedentary behaviour and the independent and combined effects it has with cardiometabolic risk markers.

Therefore, the following study was conducted to explore the effects that dietary intake had on cardiometabolic risk markers when sitting was prolonged or when sitting was interrupted. The findings from this study complement the findings from the observational studies and provide novel evidence on the role that dietary intake

plays in the sedentary behaviour and cardiometabolic health relationship in a controlled, clinical setting.

The following study has been published as a manuscript in the *Journal of Science and Medicine in Sport* (Impact factor; 3.756) and has been formatted according to their requirements. The citation for the manuscript is: *Fletcher EA, Salmon J, McNaughton SA, Orellana L, Wadley GD, Bruce C, Dempsey PC, Lacy KE, Dunstan DW. Effects of breaking up sitting on adolescents' postprandial glucose after consuming meals varying in energy: a cross-over randomised trial. Journal of Science and Medicine in Sport (2017) (in press)*. The manuscript supplementary files can be found directly after the manuscript in *Section 7.3*. The 'Authorship Statement' for this manuscript and a copy of the 'Permission Statement to Publish' this manuscript are contained in Appendix F.1 and Appendix F.2, respectively.

Findings from this article were presented at the International Society for Physical Activity and Health in Bangkok, Thailand in November 2016 (abstract available in Appendix F.3) and at the NHMRC Centre for Research Excellent in sitting and chronic conditions symposium at Baker Heart and Diabetes Institute, Melbourne in December 2016.

For the following study, the candidate was actively involved in all aspects of the study design and data collection process, as well as the manuscript process. A specific list of the candidate's contributions to the study are outlined below:

Contributions to the study design and data collection

- Substantially contributed to the design and protocol of the study

- Drafted and submitted the National Ethics Application Form to Deakin University Human Research Ethics Committee (Ethics approval in Appendix F.4)
- Drafted the Plain Language Statement and Consent form used in the study (Appendix F.5)
- Developed all study materials including the main surveys (Appendix F.6 and Appendix F.7), 1-day food diary (Appendix F.8), checklist survey (Appendix F.9, food intake records (Appendix F.10), study instructions for participants and study protocol.
- Designed the recruitment materials including flyers, brochures and advertising materials (Appendix F.11).
- Actively recruited all adolescent participants into the study.
- Scheduled, attended and collected the data for all research visits at the participant's home and at the laboratory
- Obtained accreditation to safely fit and remove the continuous glucose monitoring systems to participants
- Created and distributed individualised report cards for all participants (Appendix F.12)
- Processed the data collected, including the continuous glucose monitoring system data

Contributions to the manuscript

The candidate contributed to all aspects of the manuscript process, with the author contributions to the manuscript as follows: Fletcher, Salmon, McNaughton, Bruce, Wadley, Lacy and Dunstan conceived and designed the study; Fletcher collected the data and calculated the main variables used in the analyses; Fletcher and Orellana

performed the main analyses; Fletcher wrote the initial draft of the manuscript; and Fletcher, Salmon, McNaughton, Bruce, Wadley, Dempsey, Orellana, Lacy and Dunstan provided critical edits to the manuscript. All authors approved the final manuscript.

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Original research

Effects of breaking up sitting on adolescents' postprandial glucose after consuming meals varying in energy: a cross-over randomised trial

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ABSTRACT

Objectives: To explore the impact of uninterrupted sitting versus sitting with resistance-type activity breaks on adolescents' postprandial glucose responses while consuming a diet varying in energy.**Design:** Cross-over randomised trial.**Methods:** Thirteen healthy participants (16.4 ± 1.3 years) completed a four-treatment cross-over trial: (1) uninterrupted sitting + high-energy diet; (2) sitting with breaks + high-energy diet; (3) uninterrupted sitting + standard-energy diet; and (4) sitting with breaks + standard-energy diet. For all four conditions, two identical meals were consumed; at 0 h and 3 h. A continuous glucose monitoring system (CGM) recorded interstitial glucose concentrations every five minutes. Linear mixed models examined differences in glucose positive incremental area under the curve (iAUC) and total AUC between the sitting and diet conditions for the first meal, second meal and entire trial period.**Results:** Compared to the uninterrupted sitting conditions, the breaks condition elicited a 36.0 mmol/L/h (95%CI 6.6–65.5) and 35.9 mmol/L/h (95%CI 6.6–65.5) lower iAUC response after the first and second meal, respectively, but not for the entire trial period or for total AUC. Compared to the standard-energy diet, the high-energy diet elicited a 55.0 mmol/L/h (95%CI 25.8–84.2) and 75.7 mmol/L/h (95%CI 8.6–142.7) higher iAUC response after the first meal and entire trial, respectively. Similar response to the high-energy diet were observed for total AUC.**Conclusions:** According to iAUC, interrupting sitting had a significant effect on lowering postprandial glucose for both dietary conditions, however, it was not significant when examining total AUC. Larger studies are needed to confirm these findings.**Clinical Trial Registration Number:** ACTRN12615001145594.© 2017 Sports Medicine Australia. Published by Elsevier Ltd. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Abbreviations: AUC, Area under the curve; CGM, Continuous glucose monitoring system.

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1. Introduction

Chronic health conditions thought to develop during late adulthood, such as metabolic syndrome and type 2 diabetes, are now occurring at a younger age than in previous decades.¹ Some studies in youth,² but not all,³ suggest that engaging in high volumes of sedentary time is associated with increased fasting blood glu-

cose levels and insulin resistance which are key risk factors for developing these conditions. Thus, targeting reductions in sedentary time has the potential to improve an adolescent's metabolic profile and reduce their risk of developing metabolic syndrome or type 2 diabetes.

In the last decade, acute experimental trials have reported that interrupting sitting time with brief bouts of light- or moderate-intensity walking reduces postprandial glucose and insulin levels in healthy^{4,5} and overweight/obese adults.^{6,7} Moreover, two recent randomised cross-over studies have extended these findings into younger populations.^{8,9} One study by Belcher et al. demonstrated that interrupting 3 h of continuous sitting with 3-min light-intensity walking breaks every 30 min reduced postprandial glucose by 7% and insulin levels by 32% in children aged 7–11 years.⁸ However, the other study by Saunders et al. reported no significant differences in glucose or insulin responses in children aged 10–14 years when 8 h of prolonged sitting was interrupted with 2-min light- or moderate-intensity walking breaks every 20 min.⁹

The inconsistent findings between experimental studies may be explained in part by disparities in the energy content of the meals and by differences in blood sampling intervals. In the study by Saunders et al.,⁹ although the meals were specifically developed for a young population, they may not have contained enough energy or the most suitable foods or beverages to raise glucose and insulin levels as has been demonstrated in adult studies.^{4–6} In contrast, the findings by Belcher et al.⁸ indicate that the use of meals containing a large amount of energy (e.g. a buffet style meal) may be necessary to elicit a sufficient glycaemic challenge to observe significant differences between intervention conditions. In addition, blood samples were collected at different time intervals (i.e. 30 min⁸ versus 90 min⁹), which may not have been sufficiently regular to capture the fluctuations in glucose often seen in healthy, young populations. Continuous glucose monitoring systems (CGM) are minimally invasive and assesses interstitial glucose concentrations every 5 min. This provides the opportunity to assess the various temporal changes in blood glucose levels.¹⁰ Although these devices are widely used clinically and in patients with type 1 or 2 diabetes,^{10,11} no study to date has used a CGM to examine the acute effects of interrupting sitting on glucose responses in a healthy, young population.

Given the key role that dietary intake has on glucose metabolism,¹² it is surprising that no study to date has examined the effects of interrupting prolonged sitting on postprandial glucose when the dietary component is manipulated. Therefore, the aim of this pilot study was to explore the impact of uninterrupted sitting versus sitting with activity breaks on adolescents' postprandial glucose responses after consuming a diet varying in energy (e.g. high-energy versus standard-energy) across three time periods; (1) after the first meal; (2) after the second meal; (3) and the entire trial period. While we anticipated that the high-energy diet would elicit increased glucose responses overall, we hypothesised that the activity breaks would significantly attenuate postprandial glucose after the first meal, second meal and the entire trial period when compared to uninterrupted sitting for both diet conditions.

2. Methods

Ethical approval was obtained by Deakin University Human Research Ethics Committee in April 2015 (#2015-039). Study participants were recruited between September and December 2015 via flyers, local newspaper advertisement and word of mouth in Melbourne, Australia. Eligible participants were aged between 14 and 17 years, in good general health, not on any glucose lowering medications and had no dietary allergies. Eligibility criteria were assessed via phone screening interview with the participant or

their parent. The recruitment stages are shown in Fig. S1. Written parental and participant consent was provided by 16 participants, with 15 participants completing the four trial conditions over the school summer holidays (November 2015 to January 2016). Participants who completed all four conditions received a FitBit® as compensation for their time.

The study was an acute, cross-over factorial randomised trial involving four experimental conditions each performed over a 6-h period: (1) uninterrupted sitting and high-energy diet; (2) interrupted sitting with 2-min activity breaks every 18-min and high-energy diet; (3) uninterrupted sitting and standard-energy diet; and (4) interrupted sitting with 2-min activity breaks every 18-min and standard-energy diet. For each condition, participants received two meals, consumed at 0 and 3 h. The 2-min activity breaks involved body-weight resistance exercises and included 30-s half squats, 30-s calf raises, 30-s knee lifts and 30-s step-ups. Resistance-type activities were selected as resistance-type activities utilises larger muscle groups,¹³ and thus may promote an increased energy expenditure and higher glucose uptake. The study protocol and an example of one of the experimental conditions is shown in Fig. 1.

After enrolment, each participant was assigned one of the pre-determined condition orders using a sequence which was randomly computer-generated by a research assistant. Participants were kept blinded to the order in which they completed each condition until the morning of each visit. Since moderate-intensity physical activity has been shown to have no residual effects on plasma glucose past a 17-h period,¹⁴ a one-day minimum washout period was selected between trial conditions. The 24-h prior to each condition, participants were asked to refrain from participating in any moderate-to-vigorous physical activity (MVPA). Compliance to MVPA was self-reported using a checklist completed the morning on each condition day.

For all four experimental conditions, participants arrived at the laboratory between 0730 and 0800 after a 12-h overnight fast. At the first visit only, participants' height, weight, waist circumference, and blood pressure were measured using standard procedures.^{15,16} Participants had access to a television, DVDs, sedentary video games, and computer and internet services. Participants were provided with a comfortable chair throughout the trial and were instructed to sit upright and minimise any movement when not performing the activity breaks. If participants needed to use the lavatory, they were escorted in a wheelchair. Research staff directly supervised the participants at all times to ensure compliance with the protocol. For descriptive purposes, participants completed a questionnaire about their usual MVPA¹⁷ and sedentary behaviours levels¹⁸ on the first condition day, and completed a 1-day food diary of food, beverages and medications consumed the day before each of the four experimental conditions. The food diaries were collected by researchers the morning of each condition to ensure compliance to the 12-h overnight fast and to calculate usual energy and macronutrient intakes.

Prior to the first and third condition, two researchers visited the participant's home to insert a CGM (Medtronic iPro2™, Northridge, USA) on the right side of the participant's lower back. After the CGM was inserted, an adhesive, waterproof strip was placed over the top of the CGM to minimise movement of the sensor and to allow participants to shower. Due to the CGM recorder having a battery life of seven days, a new sensor was inserted prior to the third trial condition to be worn during the final two conditions. To calibrate the CGM, two capillary blood glucose samples were taken from each participant by the researchers during the two home visits, and three during the trial days. The capillary blood samples were used to calibrate the CGM at the time of downloading the data. At the home visits, participants also received four standardised dinners to consume the night prior to each of the trial conditions. The

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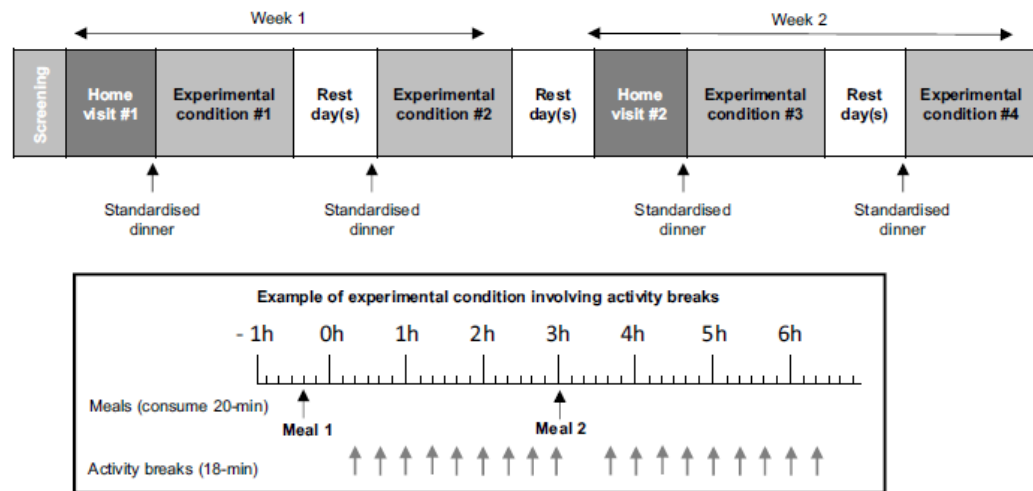


Fig. 1. Study design and experimental protocol.

standardised dinner contained 2874 kJ and included a 400 g frozen lasagne, 30 g wholemeal bread and 50 g of mixed vegetables. Participants were instructed to consume the entire meal at 1900, within 20 min and not to consume any other food or beverages except for water. Compliance to the dinner protocol was reported to the researchers the following morning.

The contents of the high-energy and standard-energy diets are described in the online supplement file (Table S1). During each of the trial conditions, participants received two high-energy meals or two standard-energy meals; the first meal was consumed at the start of the trial (between 0730 and 0830) and the second meal (exactly the same as the first meal) was provided 3 h after ingestion of the first meal. The energy contents of the meals were based on the estimated-energy requirements for an adolescent aged 17 years with a body mass index (BMI) of 25 kg/m² (averaged for females and males).¹⁹ In order to assess the effects of energy intake (kilojoules; kJ), both the high-energy and standard-energy diets contained similar macronutrient composition with the high-energy diet containing 70% more kilojoules than the standard-energy diet. The kilojoules and macronutrient contents of the diets were calculated using dietary analysis software (FoodWorks Version 7, Xyris Software Pty Ltd, Australia) based on standard Australian food composition tables.²⁰ Participants were instructed to consume the entire meal within 20 min. Any food or beverage left over was weighed and recorded and used to calculate the average percentage of kilojoules consumed during each experimental condition for each participant.

Information regarding the analyses on the sequences of the cross-over design and interaction effects can be found in the online supplement file. As this study was a pilot study, it was estimated that 16 participants (equates to 64 conditions completed) would be sufficient to determine the pragmatic considerations of examining an adolescent population. Using the CGM data, the primary outcome was positive incremental area under the curve (iAUC) and was calculated using the trapezoidal method²¹ (GraphPad Prism v6) across three time periods: (1) three-hours post first meal (includes eating time), using the baseline fasting glucose just prior to the consumption of the first meal as the reference point; (2) three-hours post second meal (includes eating time), using the glucose value just prior to consuming the second meal as baseline; and

(3) the entire trial time (including both the first and second meal responses), using the baseline fasting glucose just prior to the consumption of the first meal as the reference point. A secondary outcome was to examine total AUC. Similar to iAUC, total AUC was calculated using the trapezoidal method across the same three time periods, however, zero was used as the reference point for all time periods.

Linear mixed models were used to examine the effect of the sitting and diet conditions on iAUC and total AUC with period effect (i.e. the effect of the order at which the participant received the condition), sitting/breaks and diet conditions as fixed effects, and participant as a random effect. The interaction between the sitting/breaks and diet conditions was not examined as the pilot study was not powered to detect this. To account for potential differences in the percentage of meals consumed and fasting baseline glucose at each condition, these variables were adjusted for in the analyses. Pseudo effect size measure (Cohen's D) were calculated using the EffectSizeMixed SAS macro.²² Post-hoc power calculations were performed, with results in the online supplement file. Analyses were conducted in SAS version 9.3 (SAS Institute, Cary, NC) and $p < 0.05$ was considered significant.

3. Results

Out of the 16 participants who were randomised, 15 participants completed the four experimental conditions. Two participants were excluded from the analysis due to non-compliance to the study protocol, leaving 13 participants with complete data for the analyses. The baseline characteristics of the 13 participants are outlined in the online supplement file (Table S2). Overall, the mean age of participants was 16.4 ± 1.3 years (39% male) and BMI was 20.6 ± 2.5 kg/m². On average, participants spent 428 ± 144 min/day in self-reported sedentary behaviour and 43 ± 44 min/day in self-reported MVPA.

Fig. 2 summarises mean glucose responses across the four conditions and Table 1 summarises the AUCs responses across the four conditions and differences in the sitting/breaks and diet effects. Compared to the sitting conditions, the breaks conditions elicited a 36.0 mmol/L/h (95%CI 6.6–65.5) and 35.9 mmol/L/h (95%CI 8.6–63.1) significantly lower iAUC response after the first

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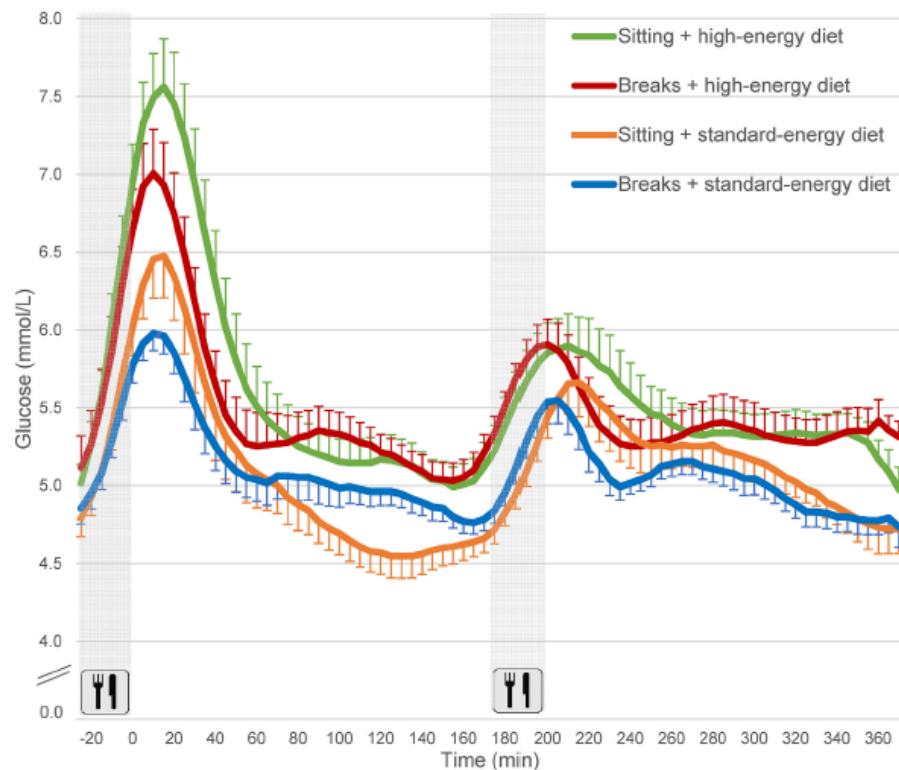


Fig. 2. Mean (SE) postprandial glucose responses across the four conditions.

Table 1
Mean \pm SE area under the curve (AUC) for all four conditions and differences (95%CI) in the effects of the sitting conditions and diet conditions.

	Mean (SE) AUCs for each condition ^a				Sitting versus breaks ^b		High-energy versus standard-energy ^b	
	(1) High-energy diet + sitting	(2) High-energy diet + breaks	(3) Standard-energy diet + sitting	(4) Standard-energy diet + breaks	Difference (95%CI)	Effect size	Difference (95%CI)	Effect size
iAUC (mmol/L/h)								
First meal	166.6 (21.8)	115.5 (19.1)	93.0 (14.1)	75.1 (12.7)	36.0 (6.6, 65.5) [*]	0.7	55.0 (25.8, 84.2) ^{**}	1.1
Second meal	105.1 (13.6)	84.3 (16.1)	116.4 (22.4)	70.9 (10.1)	35.9 (8.6, 63.1) [*]	0.8	2.5 (-25.3, 30.2)	0.1
Entire period	262.6 (41.4)	207.6 (48.2)	175.3 (30.1)	132.7 (24.4)	46.8 (-19.7, 113.4)	0.4	75.7 (8.6, 142.7) [*]	0.7
Total AUC (mmol/L/h)								
First meal	1145.4 (27.4)	1114.5 (27.3)	1021.7 (27.9)	1029.0 (27.5)	12.8 (-24.9, 50.4)	0.2	103.8 (66.6, 141.1) ^{**}	1.6
Second meal	1093.5 (23.0)	1084.9 (22.7)	1026.1 (23.5)	1004.6 (23.0)	30.9 (-36.4, 98.1)	0.2	78.2 (36.3, 112.1) ^{**}	1.1
Entire period	2205.0 (46.4)	2164.9 (46.1)	2037.8 (47.3)	2017.1 (46.5)	14.8 (-22.4, 51.9)	0.3	157.1 (89.1, 225.1) ^{**}	1.3

^a Mean and standard error (SE) for each condition estimated under a linear mixed model including participant as a random effect and period, diet, breaks, interaction diet/breaks as fixed effects.

^b Sitting/breaks and high-energy/standard energy diet mean differences (95% CI) estimated under the same model, excluding the interaction term which was not significant in the former model.

^{*} $p < 0.05$.

^{**} $p < 0.001$.

and second meal, respectively. However, this was no longer significant when examining iAUC across the entire trial period, nor for total AUC for any of the three time periods. For both the iAUC and total AUC, the breaks conditions had a moderate/high (0.4–0.7) and small effect (0.2–0.3), respectively, on lowering postprandial glucose. Relative to the standard-energy diet, the high-energy

diet elicited a 55.0 mmol/L/h (95%CI 25.8–84.2) and 75.7 mmol/L/h (95%CI 8.6–142.7) higher iAUC response after the first meal and entire trial, respectively. No significant diet effects on iAUC were observed for the second meal. Similar responses to the high-energy meal were observed for total AUC, with large effect sizes observed (1.1–1.6).

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4. Discussion

This is the first study to explore the impact of uninterrupted sitting and breaking up sitting with resistance-type activity breaks on adolescents' postprandial glucose while consuming a diet varying in energy. The findings showed that when examining the first and second meal responses separately, the activity breaks significantly attenuated the postprandial iAUC responses after both the first and second meal for both diet conditions, but did not significantly attenuate iAUC for the entire trial period, nor for when total AUC was examined. In addition, relative to the standard-energy diet, the study found that the high-energy diet elicit a higher postprandial glucose response for both iAUC and total AUC for both the sitting and breaks conditions.

Previous studies in children⁸ and adults⁴⁻⁷ have shown interrupting sitting time has beneficial metabolic effects on glycemic control. However, this is the first study to examine, whether regularly interrupting sitting time with activity breaks has beneficial effects on postprandial iAUC after consuming both high-energy and standard-energy diets. Interestingly, activity breaks significantly lowered iAUC but not total AUC. Although iAUC is most commonly used to assess acute glucose responses to meals relative to baseline (or fasting) glucose values, it excludes data that drop below baseline. We therefore calculated both iAUC and total AUC to provide a more comprehensive picture of the glycaemic responses. The varied effects sizes for these two glucose metrics is likely attributable to the different techniques involved in their calculation, but highlights the potential importance of considering which glucose parameters are examined – particularly across consecutive meals – and their physiological relevance.

In addition, while the measures used in this study do not permit conclusions about the potential mechanisms responsible for the reductions in postprandial glucose for iAUC, increases in contraction-mediated glucose uptake as a result of the activity breaks may play an important role.²³ As the activity breaks in the current study involved simple resistance exercises that can be performed with minimal equipment, these activity breaks could be easily incorporated into pedagogical approaches or even facilitated via environmental changes (e.g. height adjustable desks) in schools²⁴ and/or in the adolescents' home.

The study also observed, compared to the standard-energy diet, consuming a high-energy diet resulted in a higher postprandial glucose response (for both iAUC and total AUC) across the entire observational period, for both the uninterrupted sitting and breaks conditions. This is not surprising, given carbohydrates play a vital role in glucose metabolism²⁵ and the high-energy diet in the study contained 70% more kilojoules (and thus a greater amount of total carbohydrates) than the standard-energy meal. However, when examining the first and second meal responses separately, the study observed a significant higher postprandial glucose response for iAUC after the first meal, but not the second meal. This suggests that the first meal of the day (e.g. breakfast) may be the biggest contributor in terms of daily glycaemic responses and that glucose tolerance may improve for the subsequent meals later in the day (e.g. lunch and dinner).^{26,27} Thus, future interventions could encourage adolescents to engage in physical activity soon after consuming breakfast (e.g. by walking to school) in order to have the most beneficial effects on postprandial glucose.

A strength of this study was the examination of four conditions performed over four separate days which allowed us to explore both effects of breaking up sitting and varying the energy content of the meals on postprandial glucose responses. Another key strength was examining an adolescent population, which, to date has been an understudied population group within the sedentary behaviour field. In addition, the use of the CGM among adolescent participants was able to capture a more complete portrayal of the postprandial

glucose response (e.g. every 5 min), as opposed to collecting blood samples at hourly intervals. Lastly, the meals provided to the participants were based on whole foods, not supplements, commonly consumed by this population group.

Limitations include not measuring other glycaemic markers such as insulin, thus, limiting the ability to interpret the entire glycaemic response. Future studies using a continuous measure of glucose should also consider measuring key hormones including plasma insulin and C-peptide in order to examine potential mechanisms related to glucose-insulin kinetics. In addition, the study included both males and females in pre- and post-pubertal stages. Given the well-known effects that puberty has on glucose metabolism,²⁸ this may have increased the variability in postprandial glucose responses within and between participants. Further, the study only examined the acute glucose responses, therefore the current results cannot be extrapolated to long-term exposures. Lastly, the study was limited by a small sample size and it is unclear whether the findings would be exaggerated in youth who are overweight or obese. Thus, future studies are needed involving a larger sample size and involving overweight or obese adolescents to test the protocol in an at-risk sample.

5. Conclusion

The findings from the study demonstrated that compared to prolonged sitting, regularly interrupting sitting with simple resistance activity breaks after the first and second meals attenuated postprandial iAUC responses for both the high-energy and standard-energy diets. However, no significant breaks effects were observed for total AUC. Longer-term intervention studies are needed to confirm these findings, as well as studies examining interactions between the sitting and diet conditions in relation to cardiometabolic health risk factors.

Practical implications

- Interrupting sitting time with simple resistance activity breaks lowers incremental postprandial glucose responses to both high-energy and standard-energy diets. This suggests that breaking up prolonged sitting, particularly after consuming main meals, may be an effective intervention strategy for adolescents.
- Compared to the standard-energy diet, the high-energy diet elicited a higher positive incremental postprandial glucose response after the first meal, but not the second meal. This suggests the first meal of the day may be the biggest contributor in raising postprandial glucose levels.
- Larger and longer-term interventions are needed to confirm these findings, as well as studies examining interactions between the sitting and diet conditions in relation to cardiometabolic health risk factors.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at [10.1016/j.jsams.2017.06.002](http://dx.doi.org/10.1016/j.jsams.2017.06.002).

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7.3 Manuscript supplementary files

The supplementary file supporting the manuscript “*Effects of breaking up sitting on adolescents’ postprandial glucose after consuming meals varying in energy: a cross-over randomised trial*” can be found on the following pages.

The supplementary files include:

- Statistical analyses (extended)
- Post-hoc power analyses and results
- Table S1: Contents of the high-energy and standard-energy diets provided to participants
- Table S2: Baseline characteristics of participants
- Figure S1: Participant recruitment flow diagram

Statistical analyses (extended)

Conditions were originally allocated in balanced sequences. However, attrition due to participant drop out and/or not following the diet pre-requisites for the day before rendered only 13 participants with complete data. In this sample, treatments were not equally represented at each period, therefore treatment and period effects (i.e. the effect of the order at which the participant received the condition) could have been confounded. Therefore, the period effect was ascertained under a design in which treatment and period effects were not confounded using the following approach. From the 13 participants with complete data, all subsets of 8 participants whose treatment allocation still corresponded to two different Latin Square structures (each treatment occurs only once within each sequence and once within each period) were selected, tallying 36 subsets. For each participant, linear mixed models (LMM) including period (4 levels), condition (4 levels) and the interaction period-condition as fixed effects and participant as a random effect were performed. These models were conducted for the primary outcome (iAUC) and secondary outcome (AUC total) across the three time periods (e.g. first meal, second meal and entire period). In all models, the period effect and interaction effect were not significant ($p > 0.20$ for all outcomes and periods considered). Thus, although the treatments were not originally equally represented from the 13 participants included in the study, the non-significant effect found from the period and interaction analyses suggests there were no confounding treatment or period effects.

Post-hoc power analyses and results

Post-hoc power calculations were performed using the data collected in this study on total AUC. Using a four-period, four-sequence cross-over design, a sample size of 13 achieves 38% (76%) power at a 5% significance level when the absolute difference between the two treatment means is at 50 (80) and the within-subject standard deviation is 105 (estimated from our data).

Table S1: Contents of the high-energy and standard-energy diets provided to participants

	High-energy diet [†]	Standard-energy diet [†]
Kilojoules, (%EER)	4,244 (39.0)	2,485 (23.0)
Macronutrient, g (%)		
Carbohydrates	139.7 (55)	68.0 (53)
Fat	34.3 (30)	22.0 (31)
Protein	36.2 (15)	25.7 (16)
Contents	250g pizza, 15g cheese, 53g chocolate bar, 350ml caffeine-free soda	1 x chicken, salad sandwich (85g wholemeal bread, 35g shaved chicken, 45g tomato, 20g spinach, 40g avocado), 100g low-fat yoghurt, 50g banana, 70g apple, 15g almonds, 350ml water

EER: Estimated energy requirement

[†]Participants received two meals of either the high-energy diet or standard-energy diet. The first meal was consumed at baseline (0-hour), and the second meal (exactly the same as the first meal) three hours after finishing the first meal.

Table S2: Baseline characteristics of participants

	Total (n = 13) Mean ± SD
Age (years)	16.4 ± 1.3
Sex	
Males, n (%)	5 (39)
Females, n (%)	8 (62)
Pubertal stage	
Pubertal, n (%)	6 (46)
Post-pubertal, n (%)	7 (54)
Cardiometabolic components	
BMI, kg/m ²	20.6 ± 2.5
Waist circumference, cm	70.5 ± 6.4
Systolic BP, mm Hg	107.5 ± 8.3
Diastolic BP, mm Hg	60.0 ± 8.1
Fasting glucose, mmol/L [†]	4.8 ± 0.3
Habitual activity	
Total sedentary time, min/day	428.1 ± 143.6
MVPA, min/day	43.1 ± 44.1
Dietary intake^{††}	
Energy intake, kj	7012.5 ± 1090.1
Carbohydrate, %	56.3 ± 5.0
Protein, %	16.4 ± 5.5
Fat, %	27.3 ± 3.3

Results are expressed as mean±SD unless otherwise indicated.

[†]Based on the average fasting glucose across all four conditions.

^{††}Based on the average dietary intake from 4 x 1-day food diaries.

MVPA: moderate-to-vigorous physical activity; kj: kilojoules

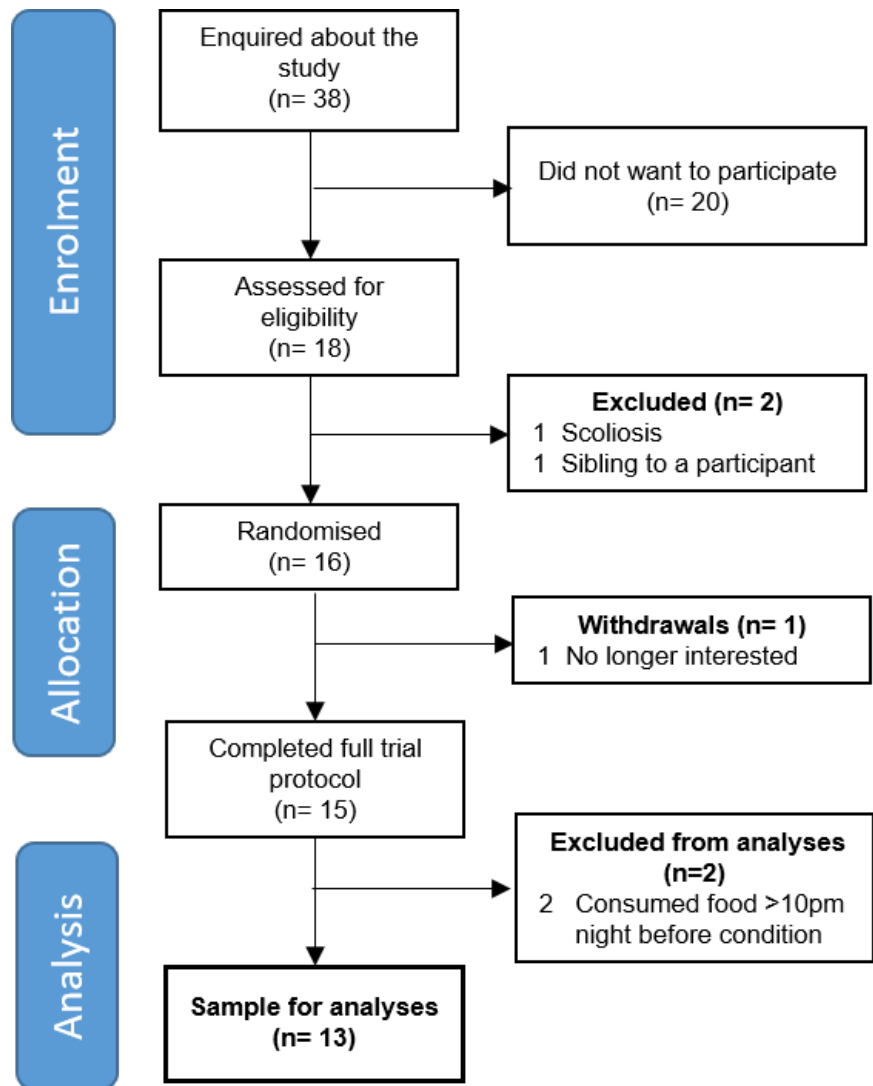


Figure S1: Participant recruitment flow diagram

7.4 Summary

The experimental study presented in Chapter Seven is the first study in the world (for any age group) to examine the combined effects of dietary intake and sitting on postprandial glucose using a continuous glucose monitoring system. Overall, the study found compared to the uninterrupted sitting conditions, the activity breaks conditions significantly attenuated postprandial glucose responses for both the high-energy and standard-energy diets. These results provide the first international insights that interrupting sitting regardless of the energy content of the meal, can reduce the impact of prolonged sitting on postprandial glucose in adolescents.

In addition to the findings presented in the manuscript in *Section 7.2*, the candidate also performed additional analyses on the thirteen participants who completed the experimental trial and complied to the study protocol (results of the additional analyses found in Appendix F.13 and Appendix F.14). Briefly, the candidate explored the effects of the sitting and diet conditions had on the first glucose peak response (e.g. the glucose response following the first meal, starting at baseline fasting glucose and finishing when the glucose returned back to baseline). Results show that compared to the uninterrupted sitting condition, the breaks conditions elicited a 0.7 mmol/L (95% CI 0.3-1.0), or 29%, lower first peak glucose response (Appendix F.13). These results concur with the findings in the manuscript, that breaking up sitting with resistance-type activity breaks is important in lowering postprandial glucose response, in particular, the first glucose peak response.

In addition, the candidate also explored differences in the percentage of meals consumed between males and female participants. The results showed significant differences in sex ($p=0.019$), with male participants consuming a higher percentage

of the high-energy diet (100, 98.4-100) [median, IQR range] compared to the standard-energy diet (97.5, 76.2-100); whereas, the female participants consumed a higher percentage of the standard-energy diet (87.7, 79.0-100) compared to the high-energy diet (76.5, 63.4-88.5) (Appendix F.14). These findings have informed a larger clinical trial involving similar conditions; that being, the diets provided to participants are tailored by sex.

CHAPTER EIGHT: DISCUSSION

CHAPTER EIGHT: DISCUSSION

8.1 Summary of main findings

As outlined in the Introduction, TV viewing, but not total volume or bouts of sedentary time, is consistently associated with cardiometabolic health in adolescents. Dietary intake could partially explain why TV viewing is consistently related to cardiometabolic health outcomes, however, very few studies have examined this. Therefore, this thesis aimed to examine the role of dietary intake in the relationship between various sedentary behaviours (e.g. TV viewing, total volume and bouts of sedentary time) and key cardiometabolic health indicators (e.g. BMI, metabolic syndrome and postprandial glucose) in adolescents.

A multidisciplinary approach was undertaken to achieve this aim. Four studies were conducted using two existing cross-sectional and longitudinal datasets and original data collected from a cross-over experimental trial. Overall, despite some significant mediation effects observed for *Study 1*^a, the combined findings from the four studies suggest that dietary intake *does not* play a major role in the relationship between sedentary behaviour and cardiometabolic health in adolescents. *Study 1* found that sugar-sweetened beverage consumption and fruit and vegetable intake only partially mediated the relationship between TV viewing and metabolic syndrome, with no

^a Study 1 – Chapter Four: Mediating effects of diet on associations of TV viewing, body mass index and metabolic syndrome in adolescents

^b Study 2 – Chapter Five: Mediating effects of diet on associations of volume and bouts of sedentary time and cardiometabolic health in adolescents

^c Study 3 – Chapter Six: Cross-sectional and prospective mediation effects of diet on associations between sedentary behaviour and body mass index in adolescents

^d Study 4 – Chapter Seven: Effects of breaking up sitting on adolescents' postprandial glucose after consuming meals varying in energy: an experimental trial

significant mediation effects observed for the relationship between TV viewing and BMI. In addition, neither *Study 2^b* and *Study 3^c* found that any of the dietary variables examined mediated the relationships between objectively-measured total sedentary time or bouts of sedentary time and BMI or metabolic syndrome, cross-sectionally (*Studies 2 and 3*) or prospectively (*Study 3*). Lastly, the findings from the experimental trial (*Study 4^d*) found compared to the prolonged sitting conditions, when sitting was interrupted with resistance-type activity breaks, the postprandial glucose response was significantly lower for both diet conditions (i.e. high-energy and standard-energy diets).

In the following sections, the key findings of the studies reported in Chapters Four to Seven will be discussed in relation to the current literature, as well as the overall strengths and limitations of this thesis. Thereafter, the public health implications and opportunities, and directions for future research will be discussed.

8.2 General discussion

8.2.1 The role of diet in the relationship between sedentary behaviour and cardiometabolic risk factors

TV viewing and cardiometabolic risk factors

In contrast to the thesis hypothesis that the consistent associations observed between TV viewing and BMI were due to an increase in dietary intake or the displacement of healthy dietary behaviours, this thesis found five elements of dietary intake did not mediate the relationship between TV viewing and BMI, either cross-sectionally or prospectively. This is consistent with a previous study in adolescents that reported

TV snacking and junk food consumption were not significant mediators of the TV viewing and BMI relationship.¹

However, when examining the role of dietary intake in the TV viewing and BMI relationship in other age groups, such as pre-school aged children^{2,3} and young adults,⁴ dietary intake appears to play a significant role. Fuller-Tyszkiewicz and colleagues examined the mediating role of dietary intake in the TV viewing and BMI relationship in 9,064 Australian pre-school aged (4-5 years) children. Significant and positive cross-sectional and prospective associations between children's TV viewing and BMI were found, with discretionary food intake (e.g. hot chips, potato chips and biscuits) and sugar-sweetened beverage consumption (e.g. juice and soft drink) partially mediating the prospective association.² Similar results have also been observed in cross-sectional studies involving middle-aged adults, where food and beverages consumed whilst watching TV and snack food consumption partially mediated the positive relationship found between TV viewing and obesity.^{4,5}

Despite the findings in Chapter Four and Six that five elements of dietary intake did not have a significant mediating effect in the TV viewing and BMI relationship, findings from *Study 1*, but not *Study 3*, demonstrated that the consumption of sugar-sweetened beverages and fruit and vegetable intake did play a small, but significant, role in the TV viewing and metabolic syndrome relationship. To summarise, *Study 1* found the significant and positive relationship between TV viewing and metabolic syndrome in U.S. adolescents was partially explained by the consumption of sugar-sweetened beverages (4.1%) and fruit and vegetable intake (8.7%). Although no other study to date (in any age group) has specifically examined the mediating role of diet in the TV viewing and metabolic syndrome relationship, cross-sectional studies

in adults have found the relationship between TV viewing time and metabolic syndrome to be independent of dietary intake.^{6,7} For example, findings from the Australian Diabetes, Obesity and Lifestyle study (AusDiab) involving 5,682 adults (aged ≥ 35 years), showed that high TV viewing time significantly increased the risk of metabolic syndrome, even after adjusting for high snack food consumption.⁷ Although the mediating role of dietary intake cannot be determined in this study, the findings suggest that the relationship between TV viewing and metabolic syndrome remain significant even when taking into account high snack food consumption.

There could be a number of reasons for the mixed findings observed between the studies included in the current thesis and previous studies that have examined dietary intake in the TV viewing, BMI and metabolic syndrome relationships. Firstly, the mixed findings could be due to the different population and age groups examined. For example, previous studies have found mediating dietary effects in young children and young adults, but not in adolescents. This could be due to measurement error, where studies have found adolescents are often less interested, less motivated and less cooperative when reporting their dietary intake compared to younger children and older adults, thus increasing reporting error.⁸ In addition, other studies have found adolescents under-report time spent in sedentary behaviors more so than adults, with weak correlations observed between reporting sedentary behaviour *versus* accelerometry-measured sedentary behaviour in adolescents (Spearman $r = 0.14$; 95% CI 0.05, 0.23).⁹

Secondly, issues relating to the survey measures cannot be discounted, since both the studies in Chapters Four and Six did not measure TV viewing concurrently with dietary intake (e.g. TV viewing data and dietary data were reported for different

days). This makes it difficult to assess how much of daily TV viewing time was also spent engaging in eating occasions. Thus, other indirect behavioural mechanisms¹⁰ such as adolescents who watch large amounts of TV and also have a poor dietary intake (but are not necessarily consuming this whilst watching TV) could explain some of the associations observed between TV viewing and adverse cardiometabolic health.

Lastly, there are numerous dietary variables that could be examined as potential mediators between the TV viewing and cardiometabolic health relationship.

Although the current thesis examined a variety of dietary variables (e.g. total energy intake, fruit and vegetable intake, frequency of consumption and intakes of sugar-sweetened beverages and discretionary foods, and dietary quality score), there are other key dietary variables, or combinations of dietary variables, that may also be important. For example, snacking whilst watching TV, frequency of consuming junk food and an unhealthy dietary score have also been associated with TV viewing¹¹ and/or adverse cardiometabolic health outcomes¹² and, thus, could potentially mediate TV viewing and cardiometabolic health relationships.

Total volume and bouts of sedentary time and cardiometabolic risk factors

Chapters Five and Six presented evidence of the role of diet in the relationships between objectively-measured total volume of sedentary time and bouts of sedentary time with BMI and metabolic syndrome. Overall, both studies found that none of the dietary variables played a significant mediating role in the relationships between total volume or bouts of sedentary time with either BMI or metabolic syndrome, when considered cross-sectionally or prospectively. As mentioned previously, these null findings could be a reflection of the sedentary time data not being collected for the

same timeframe as the dietary intake data. For example, the 2 x 24 hour dietary recalls (*Study 2*) and the food frequency questionnaire (*Study 3*) were performed on different days to when the accelerometers were worn to capture sedentary time. This makes it difficult to assess how much of the daily sedentary time was also spent engaging in eating occasions.

Currently, there is very limited evidence on the role of dietary intake in the associations between objectively-measured sedentary time and cardiometabolic risk markers in adolescents. Although there are many studies in adolescents that have measured the associations between total volume or bouts of sedentary time and various cardiometabolic health indicators,¹³⁻¹⁶ the majority of these studies have not considered the role of dietary intake. Of the limited studies available in youth that have measured all three types of data (e.g. accelerometry-measured sedentary time, dietary intake and health indicators), all have adjusted for dietary intake in the regression analyses¹⁷⁻²⁰ and not necessarily provided both the unadjusted and adjusted results when controlling for dietary intake.^{17, 19, 20}

For example, Carson and colleagues examined the associations between sedentary time with various cardiometabolic risk factors (e.g. waist circumference, systolic blood pressure, low HDL-cholesterol and C-reactive protein) in 2,527 U.S. youth aged 6-19 years.¹⁷ They found no significant associations between total volume or bouts of sedentary time and any of the cardiometabolic risk factors after adjusting for dietary percentages of fat and saturated fat, cholesterol and sodium consumed as well as other key covariates. However, by only presenting the adjusted results, it makes it difficult to determine the mediating role, or even the independent contributions, that

dietary intake may have had on the sedentary behaviour and cardiometabolic health relationship.

In contrast, Gabel and colleagues examined both the unadjusted and adjusted results in the relationships between the frequency of sedentary bouts and various cardiometabolic markers in 164 Australian children aged 7-10 years.¹⁸ Although the frequency of sedentary bouts (lasting between 5-10 minutes) was positively associated with C-reactive protein (a marker of inflammation), after adjusting for energy density (defined as the frequency of consuming energy-dense foods and drinks), the relationship was no longer significant. This suggests that dietary intake may have mediated or been a key driver in the significant association observed.

Given the lack of evidence examining objectively-measured sedentary time, dietary intake and cardiometabolic risk markers together in youth, further studies are needed to confirm the role of dietary intake in the objectively-measured sedentary time and cardiometabolic health relationship. Specific recommendations for future studies examining objectively-measured sedentary time are provided later in the discussion, as the last section of the role of dietary intake in the relationship between prolonged/interrupted sitting and cardiometabolic health will now be outlined.

Experimental evidence of the acute effects of breaking up prolonged sitting on cardiometabolic markers

A key limitation of the studies in Chapters Four, Five and Six was that TV viewing and objectively-measured sedentary time were not assessed concurrently with dietary intake. However, the findings from the experimental cross-over study from Chapter Seven provided concurrent data on sedentary time and dietary intake, as well as postprandial glucose data. This not only helps to fill the gaps in the observational

findings, but provides key insights in understanding the role of dietary intake in real-time. Overall, the findings from Chapter Seven were similar to the observational findings in Chapters Four to Six, where dietary intake did not appear to play a major role in the relationships between prolonged or interrupted sitting time and postprandial glucose.

Although many studies have shown that interrupting sitting time is beneficial for postprandial glucose and insulin,²¹⁻²⁵ the experimental trial presented in Chapter Seven is the first study to show interrupting sitting time has a beneficial impact on positive postprandial glucose, regardless of whether the participants are consuming an ‘unhealthy’ diet (e.g. high-energy) or ‘healthy’ diet (e.g. standard-energy).

However, it is important to note, the activity breaks were only beneficial in lowering positive postprandial glucose, and not when total postprandial glucose area under the curve was examined. As previously mentioned in Chapter Seven, these mixed findings between the two glucose metrics are most likely attributable to the different techniques involved in their calculation. Thus, it is important for future studies examining consecutive meals across the day to carefully consider which glucose parameters are examined.

The experimental trial in Chapter Seven also found that, relative to the standard-energy diet, there was a 36% higher positive glucose area under the curve response after consuming the first high-energy meal, but not the second. This suggests that although interrupting sitting is important for lowering postprandial glucose, dietary intake is the major determinant of glucose and energy metabolism across the day. In addition, since only the first high-energy meal significantly increased the glucose area under the curve compared to the second high-energy meal, this suggests the first

meal of the day (i.e. breakfast) may be a more important time to break up sitting compared to after consuming other main meals (e.g. lunch and dinner).

Although no other study to date has specifically explored how changing the energy content of the diet influences cardiometabolic markers when sitting is prolonged or when sitting is interrupted with resistance-type activity breaks, similar study designs in adults have explored other dietary factors. Recently, in a randomised cross-over trial, the effects of the glycaemic index of a diet (high *versus* low) on postprandial glucose and insulin responses when 4-hours of sitting was interrupted with moderate-intensity activity were explored in 14 male adults (mean \pm SD age 22.1 ± 1.2 years).²⁶ Compared to the sitting condition, breaking up prolonged sitting following either a high or low glycaemic index breakfast significantly attenuated the postprandial glucose responses by 36% and 30%, respectively. This suggests that breaking up sitting is beneficial regardless of the glycaemic index of the diet consumed.

In addition, another study by Bailey and colleagues²³ also explored the differences in *ab libitum* energy intake, and appetite and gut hormone concentrations when 5 hours of sitting was broken up with light-intensity or moderate-intensity walking in 13 healthy female adults. The authors found no significant differences in *ad libitum* energy intake between conditions when sitting was broken up with light or moderate-intensity walking and that breaking up sitting time did not affect either appetite or gut hormone concentrations. These initial findings suggest that in an acute setting, breaking up sitting time may not influence appetite or gut hormone, thus may not promote an increase in energy intake.

Interestingly to note, it appears the intensity, or type of activity breaks performed during the interrupted sitting conditions had similar beneficial effects in lowering postprandial glucose responses. For example, even though the experimental trial in Chapter Seven involved 2-minute resistance-type activity breaks performed every 18 minutes (e.g. calf raises, body squats, knee raises) and the study conducted by Bailey et al²⁶ involved 2-minute moderate-intensity activity breaks performed every 20 minutes (e.g more aerobic-based), both studies found the activity breaks had beneficial effects on lowering postprandial glucose, irrespective of the dietary condition. Similar findings have also been observed in other experimental trials in overweight adults²¹ and adults with type 2 diabetes.²² Although these studies did not manipulate the dietary condition (e.g. all participants consumed the same standardised diet for all conditions), the studies found light-intensity²¹, moderate-intensity,²¹ and resistance-type activity breaks²² had beneficial effects in lowering postprandial glucose. Collectively, the findings suggest that interrupting sedentary time with any form of activity can significantly attenuate acute postprandial glucose responses.

In addition, substitution analyses in adults have also shown that replacing sedentary time with light and moderate physical activity reduces mortality risk.^{27, 28} For example, Fishman et al²⁸ found replacing 30 minutes of sedentary time with light-intensity activity or moderate-intensity activity could result in a 20-51% reduction in mortality risk in American adults aged 50 to 79 years. Further studies are needed using substitution analyses techniques in studies involving adolescents to investigate whether these beneficial health outcomes are evident in a younger population.

Summary

In summary, despite having observed some significant cross-sectional mediation effects between TV viewing and metabolic syndrome, the combined findings from the observational studies and experimental trial suggest that the dietary variables measured in this thesis do not appear to play a major role in the sedentary behaviour and cardiometabolic risk factor relationships examined. However, when comparing these findings to previous studies, it appears the literature is mixed on whether dietary intake does play a key role. Further research using mediation analyses to determine the contribution dietary intake has in the sedentary behaviour and cardiometabolic health relationship is urgently needed. Additionally, given that very few studies in youth have concurrently measured both sedentary behaviour *and* dietary intake using objective (e.g. accelerometry) or gold-standard measures (e.g. 24-hour recalls), further studies are also needed in this area (see *Section 8.5* for further discussion).

8.2.2 Associations between various sedentary behaviours and cardiometabolic health outcomes

In addition to examining the role of dietary intake in the sedentary behaviour and cardiometabolic health relationship, the current thesis also explored the independent associations between various ‘types’, or measures, of sedentary behaviours (e.g. TV viewing, and total volume and bouts of sedentary time) and cardiometabolic risk markers in adolescents. Overall, the findings from the thesis were mixed, indicating that the associations were dependent on the sedentary behaviour measure used. For example, this thesis observed significant, positive relationships when examining TV viewing time with BMI and metabolic syndrome in adolescents, however, when examining total and bouts of sedentary time, no significant associations were found

with either BMI or metabolic syndrome, after adjusting for moderate-to-vigorous physical activity. The mixed findings from this thesis concur with the literature presented in Chapter One, where more consistent relationships are often observed when examining TV viewing time with health indicators, compared to examining overall sedentary time in a young population group.^{29, 30}

To date, only one other study has specifically explored the differences in associations between TV viewing *and* overall sedentary time with cardiometabolic health in youth.³¹ Findings from The 2008 Health Survey for England involving 4,469 children (aged 5-15 years) found TV viewing time, but not objectively-measured total sedentary time, was consistently associated with higher levels of obesity; every hour spent watching TV increased the risk of obesity by 42%.³¹ It was hypothesised that the differences in findings between TV viewing and total sedentary time could be attributed to an increase in high-calorie snack food consumption when watching TV. However, the findings from this thesis suggest dietary intake may not play a significant mediating role in the relationship between TV viewing and BMI. Thus, further studies are warranted in order to better understand the role of dietary intake in the sedentary behaviour and cardiometabolic relationship.

8.3 Strengths and limitations

The current thesis has a number of key strengths and limitations that need to be considered. Firstly, the primary strength of this thesis is that it included analyses using large observational datasets from two countries and a novel four-treatment, cross-over trial. This allowed the candidate to examine the associations at a population level, both cross-sectionally and prospectively, as well as the combined effects of dietary intake and sitting on postprandial glucose in a controlled acute

setting. Secondly, the experimental trial in Chapter Seven is the first in the world (for any age group) to examine four different experimental conditions for sitting and dietary intake and to show interrupting sitting can reduce postprandial glucose responses in adolescents after consuming a high-energy or standard-energy diet. It is also one of the first studies to examine the effects of ‘real food’ (as opposed to a standardised drink) on postprandial glucose collected every 5 minutes using a continuous glucose monitoring system.

Other strengths of this thesis include examining a combination of self-reported and objective measures of sedentary behaviour. This allowed the candidate to explore whether the differences in associations between various sedentary behaviours and cardiometabolic risk markers could partially be explained by dietary intake. It also allowed the candidate to explore the unique contributions that the various sedentary behaviours had with both dietary intake and cardiometabolic risk markers. The unique contributions are of particular importance, given that the literature on objectively-measured total sedentary time and bouts of sedentary time is an understudied area in the pediatric population. A range of dietary variables were also assessed using different dietary measures such as the ‘gold-standard’ 2 x 24 hour dietary recalls and a food frequency questionnaire. This allowed the quantity and quality of dietary intake to be examined and also the frequency of food and beverage consumption over a specific time period. Lastly, all of the observational studies included in this thesis controlled for a variety of well-known confounders, including age, sex, moderate-to-vigorous physical activity, socio-economic status, ethnicity, and pubertal status. Controlling for confounding variables improves the robustness of the results reported in this thesis.

This thesis also has a number of limitations. A key limitation is that the data presented in Chapter Four, Five and Six were collected more than a decade ago, thus the sedentary behaviour and dietary intakes observed may not be reflective of the behaviours evident today. For example, advancing technology over the past decade has introduced watching TV on many portable devices (e.g. tablets, phones and laptops)³²; thus, ‘TV viewing’ a decade ago would be not be the same as ‘TV viewing’ today. In addition, the consumption of some new sugar-sweetened beverages (e.g. energy drinks and sport drinks) has increased over the last decade,³³ and these now-popular sugar-sweetened beverages are underrepresented in the studies included the current thesis. Another key limitation is the measurement error associated with self-reporting dietary intake. This is a common and often unavoidable limitation for observational studies when assessing dietary intake, especially total energy intake.³⁴ Thus, under-reporting or misreporting of dietary intake for the studies presented in Chapter Four to Six may have occurred, and potentially biased the results.

Another potential limitation is the aggregation of cardiometabolic risk markers into ‘metabolic syndrome’ in Chapters Four and Five. This can be problematic for an adolescent population as certain cardiometabolic risk markers, such as insulin, lipids and blood pressure, naturally change during adolescence due to pubertal development.³⁵ Thus, an adolescence going through puberty can be misclassified as having the metabolic syndrome. To minimise this, Chapters Four and Five used a metabolic syndrome diagnostic criteria specific for adolescents (Appendix A)³⁶ and controlled for pubertal status in the analyses. Another limitation of using the metabolic syndrome in adolescents is examining associations with small variability. In Chapters Four and Five, adolescents that were classified as having metabolic

syndrome ranged from 6.7-9.1%. Due to the small variability in data, this can preclude significant associations to be observed.

Other limitations of this thesis include only examining data from U.S. or Australian adolescents aged 12-19 years, thus the findings may not be generalisable to other population or age groups. Further, although the studies presented in Chapters Four and Five involved a large representative sample, the studies had cross-sectional designs which limited the ability to determine a causal relationship. Lastly, the experimental study in Chapter Seven was limited by a small sample size and did not measure other glycaemic markers such as insulin.

8.4 Public health implications

Individually and collectively, the results from this thesis not only make an important contribution to the sedentary behaviour literature in youth, but provide important public health messages. Although it was hypothesised that the consistent associations observed between TV viewing and BMI may be partially explained by dietary intake, the findings from this thesis showed that the dietary variables examined did not play a major mediating role. This suggests that TV viewing and dietary intake may be two independent contributors to the increased prevalence of obesity and metabolic syndrome in adolescents. Future public health strategies, which independently target reductions in TV viewing and improvements in dietary intake, may have a greater impact on adolescent health than strategies which target these behaviours combined.

In addition, the findings presented in Chapter Seven showed promising results that interrupting sitting time with resistance-type activities after consuming either the high-energy or standard-energy diet reduces postprandial glucose in healthy-weight adolescents. Although further trials with larger samples are needed to replicate these

findings, the benefits relating to the resistance-type activity breaks following both diets suggest the timing of interrupting sitting may be an important factor to consider. Currently, the sedentary behaviour guidelines for adolescents recommend breaking up long periods of sitting as often as possible (as well as recommending limiting screen time to ≤ 2 hour/day). However, there are no specific guidelines on ‘safe’ levels of sitting time. Although further evidence is needed, future sedentary behaviour guidelines could potentially include specific details about the timing of interrupting sitting (e.g. following main meals) and the frequency of breaks (e.g. every 18-minutes, 30-minutes, etc.) in order to maximise the health benefits of interrupting sitting time on postprandial glucose.

8.5 Directions for future research

The findings from this thesis have highlighted a number of potential areas for future research. These have been briefly discussed in the included manuscripts, however, more detailed recommendations are outlined below specifically for observational studies, experimental studies and interventions.

8.5.1 Observational studies

As previously discussed, the mixed findings reported in this thesis and in previous literature on whether dietary intake plays a mediating role in the sedentary behaviour and cardiometabolic health relationship could be reflective of issues relating to measurement error – where the sedentary behaviour measure is not matched with the dietary measure. Assessing these behaviours separately makes it difficult to assess how much of the sedentary behaviour spent throughout the day is also spent engaging in eating occasions. Future studies may benefit from using ‘time-use surveys’ where participants are asked to record and account all of their time (in short

episodes) over the course of one or more days, and thus may provide useful information on a range of activities and whether food/beverages were also consumed.³⁷

Emerging technologies that concurrently assess time spent in sedentary behaviour and dietary intake could also provide new insights into the mediating effects of dietary intake. For example, wearable cameras^{38, 39} and smartphone apps⁴⁰⁻⁴³ have become popular tools for measuring sedentary behaviour and dietary intake in real time and are able to capture these behaviours simultaneously. Further, Real-Time Locating Systems (RTLS: a relatively new device that automatically tracks the location of people in real time, including indoors) may provide important information about the location of participants in their natural environments (e.g. at home in the lounge room, in the backyard playing, or at school in the classroom, etc.).⁴⁴ The RTLS may also provide important information about the location of activities performed if used in conjunction with physical activity devices.

In addition, since modern technology has introduced watching TV on many portable devices, further studies are needed which assess the various ways adolescents watch TV (e.g. at home watching TV *versus* watching TV using a smartphone or tablet at home or whilst on the move) and assessing other popular screen-based activities (e.g. sedentary video games). Further to this, capturing the amount of time spent using multiple devices at once (e.g. playing games on a phone while watching TV) will help to understand whether using multiple devices at once has similar associations with cardiometabolic health risks and dietary intake when compared to examining singular sedentary behaviours. Additionally, there are many other types of sedentary behaviours (e.g. using a computer, playing sedentary video games) that could be

explored with both dietary intake and cardiometabolic health that the current thesis could not examine. Lastly, examining the characteristics of individuals (e.g. sex, socioeconomic status, BMI) who are more likely to overeat or exhibit unhealthy dietary patterns in response to sedentary behaviour activities may help provide important information in tailoring interventions specific to individuals.

8.5.2 Experimental studies

The findings outlined in Chapter Seven suggest interrupting sitting time is beneficial for postprandial glucose even after consuming a high-energy or standard-energy diet in healthy adolescents aged 14-17 years. However, due to the small sample size, the study was not powered to detect interaction effects between sitting and diet, but the study provided important pilot data (see *Section 7.3*) for performing post-hoc analyses to determine the sample sizes that would be needed for future trials to be able to detect interaction effects between conditions. Additionally, it is also unclear whether similar findings would be observed in younger children, or whether the effects are exaggerated in youth who are overweight or obese. Further, due to the experimental trial being a pilot, only interstitial postprandial glucose was assessed via a continuous glucose monitoring system. Future studies using a continuous measure of glucose should also consider measuring key hormones such as plasma insulin and C-peptide in order to examine potential mechanisms related to glucose-insulin kinetics. Lastly, the experimental trial specifically focused on total energy intake whilst keeping the percentage of macronutrients similar for both diet conditions. However, there are many other key aspects of dietary intake that could also be examined in future trials, including the glycaemic index of the diet, changing the proportion of macronutrient intake (e.g. low carbohydrate *versus* high carbohydrate) and examining the size of the meal (whole meals *versus* snacks) that

could provide new insights in the sedentary behaviour and cardiometabolic health relationship.

8.5.3 Interventions

Following on from experimental trials, a potential avenue for future interventions at schools could be to encourage students to break up their sedentary time after consuming their main meals (e.g. by encouraging active play during their lunch time break and standing up frequently in class after lunch). In addition, the findings from the experimental trial in Chapter Seven found the first meal of the day (i.e. breakfast) was the biggest contributor in terms of daily glycaemic responses compared to the other meals consumed (i.e. lunch). Thus, encouraging students to break up their sedentary time soon after consuming breakfast (e.g. by walking to school as opposed to being driven in a car, or during school class time) may have additional beneficial effects on postprandial glucose.

8.6 Conclusion

In conclusion, the findings of this thesis suggest dietary intake does not play a key role in the sedentary behaviour and cardiometabolic health relationship. This is evident from the observational studies, where only one out of the three studies found diet to play a small significant mediating role in the TV viewing and metabolic syndrome relationship, but was not found to have any mediating effects in the relationship between TV viewing and BMI, and total/bouts of sedentary time with BMI and metabolic syndrome. In addition, findings from the experimental study also confirmed the observational studies as interrupting sitting time significantly reduced postprandial glucose irrespective of the energy content of the diet. These findings provide novel evidence that the significant associations observed between TV

viewing and adverse cardiometabolic health may not be influenced by dietary intake, and that sedentary behaviour may be an independent risk factor for cardiometabolic risk factors in adolescents.

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Appendix A:
Chapter One Supporting
Documents

Appendix A.1 Measurement of metabolic syndrome according to the International Diabetes Federation criteria¹

Table 1: Age-specific metabolic syndrome[†] cut-points and corresponding IDF percentiles for males and females aged 12-19 years¹

Age (years)	WC (cm) (83 rd)	BP (mm Hg)		HDL-C (mmol/l) (26 th)	TG (mmol/l) (89 th)	Glucose (mmol/l)
		SBP (92 nd)	DBP (97 th)			
Males						
12	85.1	121	76	1.13	1.44	5.6
13	87.0	123	78	1.10	1.48	5.6
14	88.9	125	79	1.07	1.52	5.6
15	90.5	126	81	1.04	1.56	5.6
16	91.8	128	82	1.03	1.59	5.6
17	92.7	128	83	1.03	1.62	5.6
18	93.4	129	84	1.03	1.65	5.6
19	93.8	130	85	1.03	1.68	5.6
Females						
12	72.5	121	80	1.25	1.60	5.6
13	74.2	123	82	1.25	1.53	5.6
14	75.7	125	83	1.26	1.46	5.6
15	76.8	126	84	1.26	1.44	5.6
16	77.7	128	84	1.27	1.46	5.6
17	78.5	128	85	1.27	1.53	5.6
18	79.2	129	85	1.28	1.61	5.6
19	79.8	130	85	1.29	1.68	5.6

[†]Metabolic syndrome defined as having elevated waist circumference (WC) and 2 of the remaining 4 criteria. Note that elevated WC can be assumed if body mass index values above thresholds presented in Table 2. WC= Waist circumference, BP= Blood pressure, TG= triglycerides, IDF= International Diabetes Federation.

Table 2: Age-specific Body Mass Index Cut-Points (kg/m²) and corresponding percentiles for males and females¹

Age (years)	Males (89 th)	Females (84 th)
12	26.62	26.28
13	27.42	26.96
14	28.15	27.54
15	28.77	28.05
16	29.27	28.54
17	29.63	29.02
18	29.97	29.83
19	30.00	30.00

Reference: ¹ Jolliffe CJ, Janssen I. Development of age-specific adolescent metabolic syndrome criteria that are linked to the Adult Treatment Panel III and International Diabetes Federation criteria. *J Am Coll Cardiol* 2007 49(8):891-898.

Appendix B:
Chapter Two Supporting
Documents

Appendix B.1 Authorship Statement

AUTHORSHIP STATEMENT

1. Details of publication and executive author

Title of Publication		Publication details
Is the relationship between sedentary behaviour and cardiometabolic health in adolescents independent of dietary intake? A systematic review.		Published in Obesity Reviews
Name of executive author	School/Institute/Division if based at Deakin; Organisation and address if non-Deakin	Email or phone
Elly Fletcher	Institute of Physical Activity and Nutrition, Faculty of Health	elly.fletcher@deakin.edu.au

2. Inclusion of publication in a thesis

Is it intended to include this publication in a higher degree by research (HDR) thesis?	✓ Yes / No	If Yes, please complete Section 3 If No, go straight to Section 4.
---	-----------------------	---

3. HDR thesis author's declaration

Name of HDR thesis author if different from above. (If the same, write "as above")	School/Institute/Division if based at Deakin	Thesis title
As above	As above	Sedentary behaviour and cardiometabolic health in adolescents: role of diet
If there are multiple authors, give a full description of HDR thesis author's contribution to the publication (for example, how much did you contribute to the conception of the project, the design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)		
Ms Fletcher was involved in the conception and design of the manuscript, literature review, data collection and synthesis of the systematic review, and drafted and revised the manuscript for critical intellectual content.		
<i>I declare that the above is an accurate description of my contribution to this paper, and the contributions of other authors are as described below.</i>	Signature and date	Signature Redacted by Library 10/01/2017

4. Description of all author contributions

Name and affiliation of author	Contribution(s) (for example, conception of the project, design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)
Rebecca Leech	Ms Leech was involved in the data collection and synthesis of the manuscript, and revision of the manuscript for critical intellectual content.

Sarah McNaughton	Associate Professor McNaughton was involved in the conception and design of the manuscript, and revision of the manuscript for critical intellectual content
David Dunstan	Professor Dunstan was involved in the conception and design of the manuscript, and revision of the manuscript for critical intellectual content
Kathleen Lacy	Dr Lacy was involved in the conception and design of the manuscript, and revision of the manuscript for critical intellectual content
Jo Salmon	Professor Salmon was involved in the conception and design of the manuscript, and revision of the manuscript for critical intellectual content

5. Author Declarations

I agree to be named as one of the authors of this work, and confirm:

- i. that I have met the authorship criteria set out in the Deakin University Research Conduct Policy,*
- ii. that there are no other authors according to these criteria,*
- iii. that the description in Section 4 of my contribution(s) to this publication is accurate,*
- iv. that the data on which these findings are based are stored as set out in Section 7 below.*

If this work is to form part of an HDR thesis as described in Sections 2 and 3, I further

- v. consent to the incorporation of the publication into the candidate’s HDR thesis submitted to Deakin University and, if the higher degree is awarded, the subsequent publication of the thesis by the university (subject to relevant Copyright provisions).*

Name of author	Signature*	Date
Elly Fletcher	Signature Redacted by Library	16/12/2016
Rebecca Leech	Signature Redacted by Library	16/12/2016
Sarah McNaughton	Signature Redacted by Library	17/01/2017
David Dunstan	Signature Redacted by Library	16/01/2017
Kathleen Lacy	Signature Redacted by Library	16/01/2017
Jo Salmon	Signature Redacted by Library	16/01/2017

6. Other contributor declarations

I agree to be named as a non-author contributor to this work.

Name and affiliation of contributor	Contribution	Signature* and date
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N/A		
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* If an author or contributor is unavailable or otherwise unable to sign the statement of authorship, the Head of Academic Unit may sign on their behalf, noting the reason for their unavailability, provided there is no evidence to suggest that the person would object to being named as author

7. Data storage

The original data for this project are stored in the following locations. (The locations must be within an appropriate institutional setting. If the executive author is a Deakin staff member and data are stored outside Deakin University, permission for this must be given by the Head of Academic Unit within which the executive author is based.)

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N/R – systematic review of current published literature			

This form must be retained by the executive author, within the school or institute in which they are based.

If the publication is to be included as part of an HDR thesis, a copy of this form must be included in the thesis with the publication.

Appendix B.2 Copy of permission statement to publish paper

The manuscript in Chapter Two, “*Is the relationship between sedentary behaviour and cardiometabolic health in adolescents independent of dietary intake? A systematic review*” has been made publicly available. Therefore, a copy of a permission statement to publish the paper is not required. Please see evidence below.

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- Author:** E. Fletcher, R. Leech, S. A. McNaughton, D. W. Dunstan, K. E. Lacy, J. Salmon
- Publication:** Obesity Reviews
- Publisher:** John Wiley and Sons
- Date:** Jun 22, 2015

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Appendix B.3 Conference abstract: International Society of Behavioral Nutrition and Physical Activity, Edinburgh, Scotland (2015)

IS THE RELATIONSHIP BETWEEN SEDENTARY BEHAVIOUR AND CARDIOMETABOLIC HEALTH IN ADOLESCENTS INDEPENDENT OF DIETARY INTAKE? A SYSTEMATIC REVIEW

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Objective: The aim of this systematic review was to explore whether the associations between sedentary behaviour and cardiometabolic risk markers are independent of dietary intake in adolescents aged 12-18 years.

Methods: Online databases (Medline, Global Health, PsycInfo, Web of Science and Embase) and personal libraries were searched for peer-reviewed original research articles published in English before March 2014. Eligibility criteria were: 1) participants aged 12-18 years; 2) included a measure of sedentary behaviour as an independent variable; 3) controlled for dietary intake (defined as intakes of energy, macronutrients, food or beverages) in the analyses; and 4) assessed at least one cardiometabolic risk marker as the main outcome (i.e. adiposity, blood pressure, insulin sensitivity, glucose tolerance or lipid levels) or cardiometabolic conditions. Two reviewers independently screened the full-text for eligibility and rated the methodological quality of the studies.

Results: Twenty-five studies met the inclusion criteria, with all studies considered to be of high quality (quality scores ranged from 60-84%). From the 21 studies examining sedentary behaviours and adiposity, the majority found significant associations between television viewing, total screen time and total self-reported sedentary time with markers of adiposity, independent of dietary intake. However, no consistent pattern in dietary variables was identified. Three studies examined associations between sedentary behaviour and cardiometabolic markers other than adiposity, with no significant associations observed between total screen time and blood pressure. Due to a lack of studies, there was insufficient evidence of associations between sedentary time and the remaining cardiometabolic markers or metabolic syndrome.

Conclusions: This systematic review found consistent evidence that television viewing, total screen time and self-reported sedentary time are independently associated with adiposity in adolescents, irrespective of dietary intake. However, due to the variability of dietary variables that were adjusted for in the analyses, no clear pattern was identified. Additionally, no studies used objective measures to assess sedentary time and there was limited evidence to draw conclusions regarding other cardiometabolic risk markers. Thus, future work is needed to understand the role of dietary intake in the associations between screen time behaviours, objectively-measured sedentary time and cardiometabolic risk markers in adolescents.

**Appendix B.4 Symposium abstract: School of Exercise and Nutrition Sciences
10th Research Degree Symposium, Melbourne, Australia (2014)**

SEDENTARY BEHAVIOUR, DIET & CARDIOMETABOLIC HEALTH IN
ADOLESCENTS:
A SYSTEMATIC REVIEW

Fletcher E¹, Leech R¹, McNaughton S¹, Dunstan D², Lacy K¹ and Salmon J¹

¹Deakin University, Melbourne, VIC,

²Baker IDI Heart and Diabetes Institute, Melbourne, VIC.

Background/Aim: There is strong evidence supporting the association between screen time and obesity in adolescents. However, there are mixed findings when overall sedentary time is examined, with some evidence suggesting dietary intake mediates the relationship between screen time and adverse health outcomes. Therefore, the purpose of this systematic review is to explore the role dietary intake has in the relationship between sedentary behaviour and cardiometabolic risk markers in adolescents aged 12-18 years.

Methods: Online databases (Medline, Global Health, PsycInfo, Web of Science and Embase) and personal libraries were searched for peer-reviewed original research articles published in English before March 2014. Eligibility criteria were: 1) participants aged 12-18 years; 2) included a measure of sedentary behaviour as an independent variable; 3) controlled for dietary intake (defined as food and beverages consumed) in the analyses; and 4) assessed at least one cardiometabolic risk marker as the main outcome (i.e. overweight/obesity; waist circumference; blood pressure; insulin sensitivity; glucose tolerance or lipid levels). Two reviewers independently screened the full-text for eligibility and rated the methodological quality of the studies.

Results/Conclusions: Twenty-six studies met the inclusion criteria. The findings from each study will be summarised according to whether sedentary behaviour was significantly associated with cardiometabolic risk markers, and if so, whether or not the associations were independent of dietary intake. The findings will provide an understanding of the role dietary intake plays as a potential mediator between sedentary behaviour and adverse health outcomes, and whether findings differ according to how sedentary behaviour is measured.

Appendix C:
Chapter Four Supporting
Documents

Appendix C.1 Authorship Statement

AUTHORSHIP STATEMENT

1. Details of publication and executive author

Title of Publication		Publication details
Mediating effects of dietary intake on associations of TV viewing, body mass index, and metabolic syndrome in adolescents		Published in Obesity, Science & Practice
Name of executive author	School/Institute/Division if based at Deakin; Organisation and address if non-Deakin	Email or phone
Elly Fletcher	Institute of Physical Activity and Nutrition, Faculty of Health	elly.fletcher@deakin.edu.au

2. Inclusion of publication in a thesis

Is it intended to include this publication in a higher degree by research (HDR) thesis?	✓ Yes No	If Yes, please complete Section 3 If No, go straight to Section 4.
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3. HDR thesis author's declaration

Name of HDR thesis author if different from above. (If the same, write "as above")	School/Institute/Division if based at Deakin	Thesis title
As above	As above	Sedentary behaviour and cardiometabolic health in adolescents: role of diet
If there are multiple authors, give a full description of HDR thesis author's contribution to the publication (for example, how much did you contribute to the conception of the project, the design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)		
Ms Fletcher was involved in the conception and design of the manuscript, calculated the main variables used in the analyses, performed the main analyses, wrote the initial draft of the manuscript and revised the manuscript for critical intellectual content.		
<i>I declare that the above is an accurate description of my contribution to this paper, and the contributions of other authors are as described below.</i>	Signature and date	Signature Redacted by Library 10/01/2017

4. Description of all author contributions

Name and affiliation of author	Contribution(s) (for example, conception of the project, design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)
Sarah McNaughton	Associate Professor McNaughton was involved in the conception and design of the manuscript, provided statistical guidance in calculating the dietary variables, and revised the manuscript for critical intellectual content

Kathleen Lacy	Dr Lacy was involved in the conception and design of the manuscript, and revision of the manuscript for critical intellectual content
David Dunstan	Professor Dunstan was involved in the conception and design of the manuscript, and revision of the manuscript for critical intellectual content
Valarie Carson	Dr Carson provided guidance in the analyses of the data and revised the manuscript for critical intellectual content
Jo Salmon	Professor Salmon was involved in the conception and design of the manuscript, provided statistical guidance in mediation analyses, and revised the manuscript for critical intellectual content

5. Author Declarations

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- viii. that the description in Section 4 of my contribution(s) to this publication is accurate,*
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If this work is to form part of an HDR thesis as described in Sections 2 and 3, I further

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Name of author	Signature*	Date
Elly Fletcher	Signature Redacted by Library	12/01/2017
Sarah McNaughton	Signature Redacted by Library	17/01/2017
Kathleen Lacy	Signature Redacted by Library	16/01/2017
David Dunstan	Signature Redacted by Library	16/01/2017
Valarie Carson	Signature Redacted by Library	12/01/2017
Jo Salmon	Signature Redacted by Library	16/01/2017

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Name and affiliation of contributor	Contribution	Signature* and date
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Appendix C.2 Copy of permission statement to publish paper

The manuscript in Chapter Four, “*Mediating effects of dietary intake on associations of TV viewing, body mass index and metabolic syndrome in adolescents*” has been made publicly available. Therefore, a copy of a permission statement to publish the paper is not required. Please see evidence below.

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- Author:** E. A. Fletcher, S. A. McNaughton, K. E. Lacy, D. W. Dunstan, V. Carson, J. Salmon
- Publication:** Obesity Science & Practice
- Publisher:** John Wiley and Sons
- Date:** Jul 29, 2016

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Appendix C.3 Conference abstract: International Society of Behavioral Nutrition and Physical Activity, Cape Town, South Africa (2016)

MEDIATING EFFECTS OF DIET ON THE RELATIONSHIP BETWEEN TV VIEWING WITH BMI AND METABOLIC SYNDROME IN ADOLESCENTS

Elly Fletcher¹, Sarah A. McNaughton¹, Valerie Carson², David Dunstan^{1,3}, Kathleen Lacy¹, and Jo Salmon¹

¹Centre for Physical Activity and Nutrition Research, Deakin University. ²Faculty of Physical Education and Recreation, University of Alberta. ³Baker Heart and Diabetes Institute

Purpose: To determine the mediating effects of dietary intake on the relationship between television (TV) viewing, BMI and the metabolic syndrome (metS) among a large representative sample of adolescents in the United States.

Methods: The analysis was based on adolescents (12-19 years) participating in the 2003-06 US National Health and Nutrition Examination Survey. BMI z-score was calculated using age- and sex-adjusted height and weight, and metS was calculated using age- and sex-adjusted waist circumference, systolic/diastolic blood pressure, non-high-density lipoprotein cholesterol, triglycerides and plasma glucose. Hours (per day) spent watching TV were measured via a self-reported questionnaire. Dietary intake was assessed using two 24-hour recalls. A series of mediation analyses was conducted examining five dietary mediators (total energy intake, fruit and vegetable consumption, discretionary snack consumption, sugar-sweetened beverage (SSB) consumption and diet quality) on the relationship between TV viewing with BMI and the metS.

Results/findings: In total 3,161 participants had complete BMI profiles, and 1,379 participants had complete metS data. Approximately half of the sample were males, 38% were overweight or obese and 9.1% were classified as having the metS. Overall, TV viewing was significantly related to BMI ($\beta=0.099$, $p<0.001$), and remained significant after adjusting for each dietary mediators. However, no significant mediation was found with any of the dietary mediators. TV viewing was also significantly associated with the metS ($\beta=0.163$, $p=0.46$), and remained significant after adjusting for total energy intake and discretionary snack consumption. Fruit and vegetable and SSB consumption were found to be significant mediators, explaining 4.1% and 8.7% of the relationship between TV viewing and metS, respectively.

Conclusions: Contrary to our hypothesis, this study found that five aspects of dietary intake were not a significant mediator in the TV viewing and BMI relationship. However, consumptions of fruit and vegetables and SSB were found to be significant mediators in the TV viewing and metS relationship. This highlights the complexity of the relationships between TV viewing, dietary intake and cardiometabolic health outcomes. Further research is needed using longitudinal and experimental designs, and examining whether other sedentary behaviours have similar findings.

**Appendix C.4 Symposium abstract: School of Exercise and Nutrition Sciences
11th Research Degree Symposium, Melbourne, Australia (2015)**

**TELEVISION VIEWING & CARDIOMETABOLIC HEALTH
IN ADOLESCENTS: THE ROLE OF DIET**

Fletcher E¹, McNaughton S¹, Lacy K¹, Dunstan D², Carson V³, and Salmon J¹

¹Centre for Physical Activity and Nutrition Research, Deakin University, Melbourne,
²Baker IDI Heart and Diabetes Institute, Melbourne ³Faculty of Physical Education
and Recreation, University of Alberta, Canada

Background/Aim: Currently, 25% of adolescents from Western countries are either overweight or obese. Moreover, 10% are classified as having the metabolic syndrome (MetS) – a condition characterised by a collection of risk factors that increase a person’s chance of developing type 2 diabetes and cardiovascular disease. Television viewing (TV) has shown to have positive associations with both obesity and the MetS. However, dietary intake may play an important mediating role in this relationship. Therefore, the aim of this study is to determine the mediating role of dietary intake in the relationship between TV with obesity and the MetS among adolescents.

Methods: The results are based on adolescents (12-19 years) participating in the National Health and Nutrition Examination Survey (NHANES) in 2005/06. BMI z-score was calculated using age- and sex-adjusted height and weight, and MetS was calculated using age- and sex-adjusted waist circumference, systolic and diastolic blood pressure, non-high-density lipoprotein cholesterol, triglycerides and glucose. Hours spent in TV were measured via a self-reported questionnaire. Dietary intake was assessed using 2 x 24-hour recalls. A series of mediation analyses were conducted examining various dietary mediators (e.g. total energy intake, fruit and vegetable consumption, discretionary snacks, sugar-sweetened beverages and dietary quality) in the relationship between TV with BMI z-score and MetS.

Results/Conclusion: Of the 880 adolescents examined, 38% were overweight or obese and 10% had the MetS. The findings from the mediation analyses will help inform whether dietary intake plays an important role in the relationship between TV and BMI and the MetS

Appendix D:
Chapter Five Supporting
Documents

Appendix D.1 Authorship Statement

AUTHORSHIP STATEMENT

1. Details of publication and executive author

Title of Publication		Publication details
Does diet mediate associations of volume and bouts of sedentary time with cardiometabolic health indicators in adolescents?		Published in Obesity
Name of executive author	School/Institute/Division if based at Deakin; Organisation and address if non-Deakin	Email or phone
Elly Fletcher	Institute of Physical Activity and Nutrition, Faculty of Health	elly.fletcher@deakin.edu.au

2. Inclusion of publication in a thesis

Is it intended to include this publication in a higher degree by research (HDR) thesis?	✓ Yes / No	If Yes, please complete Section 3 If No, go straight to Section 4.
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3. HDR thesis author's declaration

Name of HDR thesis author if different from above. (If the same, write "as above")	School/Institute/Division if based at Deakin	Thesis title
As above	As above	Sedentary behaviour and cardiometabolic health in adolescents: role of diet
If there are multiple authors, give a full description of HDR thesis author's contribution to the publication (for example, how much did you contribute to the conception of the project, the design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)		
Ms Fletcher was involved in the conception and design of the manuscript, calculated the main variables used in the analyses, performed the main analyses, wrote the initial draft of the manuscript and revised the manuscript for critical intellectual content.		
<i>I declare that the above is an accurate description of my contribution to this paper, and the contributions of other authors are as described below.</i>	Signature and date	Signature Redacted by Library 10/01/2017

4. Description of all author contributions

Name and affiliation of author	Contribution(s) (for example, conception of the project, design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)
--------------------------------	---

Valarie Carson	Dr Carson provided statistical guidance in calculating the accelerometer-derived variables, and revised the manuscript for critical intellectual content
Sarah McNaughton	Associate Professor McNaughton was involved in the conception and design of the manuscript, provided statistical guidance in calculating the dietary variables, and revised the manuscript for critical intellectual content
David Dunstan	Professor Dunstan was involved in the conception and design of the manuscript, and revision of the manuscript for critical intellectual content
Genevieve Healy	Associate Professor Healy provided statistical guidance and revised the manuscript for critical intellectual content
Jo Salmon	Professor Salmon was involved in the conception and design of the manuscript, provided statistical guidance in mediation analyses, and revised the manuscript for critical intellectual content

5. Author Declarations

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Sarah McNaughton	Signature Redacted by Library	17/01/2017
David Dunstan	Signature Redacted by Library	16/01/2017
Genevieve Healy	Signature Redacted by Library	09/01/2017

Jo Salmon	-	16/01/2017
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Type of use	Dissertation/Thesis
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Format	Print and electronic
Portion	Full article
Will you be translating?	No
Title of your thesis / dissertation	Sedentary behaviour and cardiometabolic health: role of diet

Appendix D.3 Conference poster: International Society of Behavioral Nutrition and Physical Activity, Cape Town, South Africa (2016)

Associations of sedentary time with indicators of cardiometabolic health in adolescents: Are they mediated by diet?

Elly Fletcher*, Valerie Carson, Sarah McNaughton, David Dunstan, Genevieve Healy, Jo Salmon
Institute for Physical Activity and Nutrition, Deakin University.

INTRODUCTION

- ❖ In adolescents, associations between objectively-measured sedentary time (ST) with BMI and metabolic syndrome (MetS) are inconsistent.
- ❖ Dietary intake associated with TV viewing & plays a key role in energy metabolism
- ❖ Dietary intake could explain the consistent findings.

AIM

- ❖ Examine whether dietary intake mediates the associations between ST (volume & bouts) with BMI and MetS in adolescents (12-19y) participating in the 2003-06 NHANES.

METHODS

- ❖ zBMI and MetS were calculated using age- and sex-specific criteria.
- ❖ Data from an ActiGraph accelerometer (worn 7 days) was used to derive total ST and usual sedentary bout length.
- ❖ Diet was assessed using 2x24h dietary recalls. The mediators examined were: total energy intake (EI), fruit/vegetable intake, discretionary snacking, sugar-sweetened beverages (SSB) and diet quality (HEI2010).
- ❖ Using McKinnon method, mediation analyses assessed whether each diet variable mediated the association between ST (total & usual bout duration) with zBMI and MetS
- ❖ All models adjusted for age, sex, ethnicity, SES, diet intake under-reporters and MVPA.

RESULTS

Baseline characteristics

- ❖ zBMI sample (n= 1,797); MetS sample (n= 812)
- ❖ 35% participants (age 15.1y) were overweight/obese and 6.7% had MetS
- ❖ Mean total ST was 7.0 hr/d and average sedentary bout was 9.1 min/d
- ❖ Mean total EI was 2,230kcal and fruit/veg intake was 1.8 serves

Main findings

- ❖ No significant associations observed between ST (total or bouts) with any diet variable for both zBMI and MetS sample (a-path)
- ❖ Significant associations observed between total EI and snacks with zBMI and total EI and fruit/veg with MetS (b-path)
- ❖ No significant associations observed between ST (total or bouts) with zBMI or MetS (c-path)
- ❖ ST and zBMI adversely associated after taking into account SSB and fruit/veg intake (c'-path)
- ❖ No dietary variables significantly mediated the ST and BMI or ST and MetS relationship (a*b; mediators)

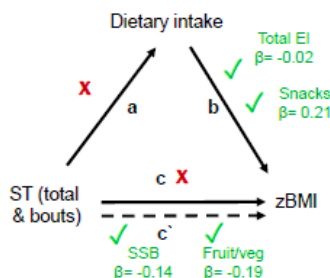


Figure 1: Mediation pathways between sedentary time, diet and zBMI (n=1,797)

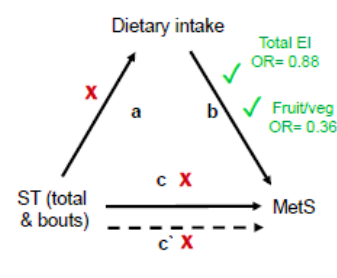


Figure 2: Mediation pathways between sedentary time, diet and MetS (n=812)

CONCLUSIONS

- ❖ No associations observed between ST with zBMI or MetS, and none of the five dietary elements mediated or masked the relationship.
- ❖ Unlike TV viewing, ST does not appear to be related to dietary intake.
- ❖ Intervention programs need to address total ST differently compared to certain sedentary behaviours (e.g. TV viewing)

*Corresponding author:
Elly Fletcher, Deakin University, elly.fletcher@deakin.edu.au
deakin.edu.au/research/ipan Deakin University CRICOS Provider Code: 00113B



Appendix E:

Chapter Six Supporting

Documents

Appendix E.1 Authorship Statement

AUTHORSHIP STATEMENT

1. Details of publication and executive author

Title of Publication		Publication details
Cross-sectional and prospective mediating effects of dietary intake on the relationship between sedentary behaviour and body mass index in adolescents		BMC Public Health
Name of executive author	School/Institute/Division if based at Deakin; Organisation and address if non-Deakin	Email or phone
Elly Fletcher	Institute of Physical Activity and Nutrition, Faculty of Health	elly.fletcher@deakin.edu.au

2. Inclusion of publication in a thesis

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As above	As above	Sedentary behaviour and cardiometabolic health in adolescents: role of diet
If there are multiple authors, give a full description of HDR thesis author's contribution to the publication (for example, how much did you contribute to the conception of the project, the design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)		
Ms Fletcher was involved in the conception and design of the manuscript, calculated the main variables used in the analyses, performed the main analyses, wrote the initial draft of the manuscript and revised the manuscript for critical intellectual content.		
<i>I declare that the above is an accurate description of my contribution to this paper, and the contributions of other authors are as described below.</i>	Signature and date	Signature Redacted by Library 10/01/2017

4. Description of all author contributions

Name and affiliation of author	Contribution(s) (for example, conception of the project, design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)
Karen Lamb	Dr Lamb was involved in the conception and design of the manuscript, advised the appropriate statistical methods to use regarding the mediation analyses, and revised the manuscript for critical intellectual content

Sarah McNaughton	Associate Professor McNaughton was involved in the conception and design of the manuscript, and revised the manuscript for critical intellectual content
Sarah Garnett	Associate Professor Garnett was a Chief Investigator of the study, was involved in the conception and design of the manuscript, and revised the manuscript for critical intellectual content
David Dunstan	Professor Dunstan was involved in the conception and design of the manuscript, and revision of the manuscript for critical intellectual content
Louise Baur	Professor Baur was a Chief Investigator of the study, was involved in the conception and design of the manuscript, and revised the manuscript for critical intellectual content
Jo Salmon	Professor Salmon was a Chief Investigator of the study, was involved in the conception and design of the manuscript, and revised the manuscript for critical intellectual content

5. Author Declarations

I agree to be named as one of the authors of this work, and confirm:

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xvii. that there are no other authors according to these criteria,

xviii. that the description in Section 4 of my contribution(s) to this publication is accurate,

xix. that the data on which these findings are based are stored as set out in Section 7 below.

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xx. consent to the incorporation of the publication into the candidate's HDR thesis submitted to Deakin University and, if the higher degree is awarded, the subsequent publication of the thesis by the university (subject to relevant Copyright provisions).

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Elly Fletcher	Signature Redacted by Library	6/01/2017
Karen Lamb	Signature Redacted by Library	6/01/2017
Sarah McNaughton	Signature Redacted by Library	17/01/2017
Sarah Garnett	Signature Redacted by Library	10/01/2017
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Louise Baur	Signature Redacted by Library	6/01/2017
Jo Salmon	Signature Redacted by Library	16/01/2017

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N/A		

* If an author or contributor is unavailable or otherwise unable to sign the statement of authorship, the Head of Academic Unit may sign on their behalf, noting the reason for their unavailability, provided there is no evidence to suggest that the person would object to being named as author

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Paper based data (e.g. surveys, consent forms)	Sydney University Discipline of Child & Adolescent Health, The Children's Hospital at Westmead Clinical School, NSW	10/01/2017	Associate Professor Sarah Garnett
Electronic information (e.g. accelerometer data, survey data, physiological measurements)	Sydney University Discipline of Child & Adolescent Health, The Children's Hospital at Westmead Clinical School, NSW	10/01/2017	Associate Professor Sarah Garnett

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Appendix F:
Chapter Seven Supporting
Documents

Appendix F.1 Authorship Statement

AUTHORSHIP STATEMENT

1. Details of publication and executive author

Title of Publication		Publication details
Effects of breaking up sitting on adolescents' postprandial glucose after consuming meals varying in energy: a cross-over randomised trial		Journal of Science and Medicine in Sport
Name of executive author	School/Institute/Division if based at Deakin; Organisation and address if non-Deakin	Email or phone
Elly Fletcher	Institute of Physical Activity and Nutrition, Faculty of Health	elly.fletcher@deakin.edu.au

2. Inclusion of publication in a thesis

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3. HDR thesis author's declaration

Name of HDR thesis author if different from above. (If the same, write "as above")	School/Institute/Division if based at Deakin	Thesis title
As above	As above	Sedentary behaviour and cardiometabolic health in adolescents: role of diet
If there are multiple authors, give a full description of HDR thesis author's contribution to the publication (for example, how much did you contribute to the conception of the project, the design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)		
Ms Fletcher was an Associate Investigator of the study and was actively involved in both the study design and data collection process. Ms Fletcher acted as the 'Project manager' which involved recruiting the adolescent participants, scheduling the research data collection visits for all participants and follow-up appointments, fitting and removing the monitors, collecting the data at each research visit, and processing the data. Ms Fletcher was also involved in the manuscript process including calculating the main variables used in the analyses, writing the initial draft of the manuscript and revising the manuscript for critical intellectual content.		
<i>I declare that the above is an accurate description of my contribution to this paper, and the contributions of other authors are as described below.</i>	Signature and date	Signature Redacted by Library
		10/01/2017

4. Description of all author contributions

Name and affiliation of author	Contribution(s) (for example, conception of the project, design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)
--------------------------------	---

Jo Salmon	Alfred Deakin Professor Salmon was a Chief Investigator of the study, was involved in the conception and design of the study, and revised the manuscript for critical intellectual content.
Sarah McNaughton	Associate Professor McNaughton was a Chief Investigator of the study, was involved in the conception and design of the study, and revised the manuscript for critical intellectual content.
Liliana Orellana	Associate Professor Orellana performed the main analyses for the manuscript, and revised the manuscript for critical intellectual content.
Glenn Wadley	Dr Wadley was a Chief Investigator of the study, was involved in the conception and design of the study, and revised the manuscript for critical intellectual content.
Clinton Bruce	Dr Bruce was a Chief Investigator of the study, was involved in the conception and design of the study, and revised the manuscript for critical intellectual content.
Paddy Dempsey	Mr Dempsey assisted in the analyses of the manuscript, and revised the manuscript for critical intellectual content.
Kathleen Lacy	Dr Lacy was a Chief Investigator of the study, was involved in the conception and design of the study, and revised the manuscript for critical intellectual content.
David Dunstan	Professor Dunstan was a Chief Investigator of the study, was involved in the conception and design of the study, and revised the manuscript for critical intellectual content.

5. Author Declarations

I agree to be named as one of the authors of this work, and confirm:

xxi. that I have met the authorship criteria set out in the Deakin University Research Conduct Policy,

xxii. that there are no other authors according to these criteria,

xxiii. that the description in Section 4 of my contribution(s) to this publication is accurate,

xxiv. that the data on which these findings are based are stored as set out in Section 7 below.

If this work is to form part of an HDR thesis as described in Sections 2 and 3, I further

xxv. consent to the incorporation of the publication into the candidate’s HDR thesis submitted to Deakin University and, if the higher degree is awarded, the subsequent publication of the thesis by the university (subject to relevant Copyright provisions).

Name of author	Signature*	Date
Elly Fletcher	Signature Redacted by Library	10/01/2017
Jo Salmon	Signature Redacted by Library	16/01/2017
Sarah McNaughton	Signature Redacted by Library	10/01/2017
Liliana Orellana	Signature Redacted by Library	10/01/2017

Glenn Wadley	Signature Redacted by Library	10/01/2017
Clinton Bruce	Signature Redacted by Library	10/01/2017
Paddy Dempsey	Signature Redacted by Library	10/01/2017
Kathleen Lacy	Signature Redacted by Library	16/01/2017
David Dunstan	Signature Redacted by Library	16/01/2017

6. Other contributor declarations

I agree to be named as a non-author contributor to this work.

Name and affiliation of contributor	Contribution	Signature* and date

* If an author or contributor is unavailable or otherwise unable to sign the statement of authorship, the Head of Academic Unit may sign on their behalf, noting the reason for their unavailability, provided there is no evidence to suggest that the person would object to being named as author

7. Data storage

The original data for this project are stored in the following locations. (The locations must be within an appropriate institutional setting. If the executive author is a Deakin staff member and data are stored outside Deakin University, permission for this must be given by the Head of Academic Unit within which the executive author is based.)

Data format	Storage Location	Date lodged	Name of custodian if other than the executive author
Paper based data (e.g. surveys, consent forms)	In a locked filing cabinet within the School of Exercise and Nutrition Sciences, Deakin University	10/01/2017	Ms Elly Fletcher Alfred Deakin Professor Jo Salmon
Electronic information (e.g. physiological measurements, glucose data, questionnaire data)	Stored on the password protected 'Research' hard drive at Deakin University	10/01/2017	Ms Elly Fletcher Alfred Deakin Professor Jo Salmon

This form must be retained by the executive author, within the school or institute in which they are based.

If the publication is to be included as part of an HDR thesis, a copy of this form must be included in the thesis with the publication.

Appendix F.2 Copy of permission statement to publish paper

The manuscript in Chapter Seven “*Effects of breaking up sitting on adolescents’ postprandial glucose after consuming meals varying in energy: an experimental trial*” has been made publicly available. Therefore, a copy of a permission statement to publish the paper is not required. Please see evidence below.

The screenshot shows the RightsLink interface with the following details:

- Title:** Effects of breaking up sitting on adolescents’ postprandial glucose after consuming meals varying in energy: a cross-over randomised trial
- Author:** Elly A. Fletcher, Jo Salmon, Sarah A. McNaughton, Liliana Orellana, Glenn D. Wadley, Clinton Bruce, Paddy C. Dempsey, Kathleen E. Lacy, David W. Dunstan
- Publication:** Journal of Science and Medicine in Sport
- Publisher:** Elsevier
- Date:** Available online 9 June 2017
- Copyright:** © 2017 Sports Medicine Australia. Published by Elsevier Ltd.

Navigation buttons include Home, Create Account, Help, and a mail icon. A LOGIN button is also present, with a note: "If you're a copyright.com user, you can login to RightsLink using your copyright.com credentials. Already a RightsLink user or want to learn more?"

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Appendix F.3 Conference abstract: International Society for Physical Activity and Health, Bangkok, Thailand (2016)

EFFECTS OF BREAKING UP SITTING ON ADOLESCENTS' POSTPRANDIAL GLUCOSE AFTER CONSUMING MEALS VARYING IN ENERGY:
A CROSS-OVER RANDOMISED TRIAL

Elly A. Fletcher¹, Jo Salmon¹, Sarah A. McNaughton¹, Liliana Orellana², Glenn D. Wadley¹, Clinton Bruce¹, Paddy C. Dempsey³, Kathleen E. Lacy¹, and David W. Dunstan^{1,3}

¹Institute for Physical Activity and Nutrition, Deakin University. ²Biostatistics Unit, Deakin University, ³Baker Heart and Diabetes Institute.

Background: Breaking up prolonged sitting may be beneficial for glucose metabolism in adolescents. However, the impact of dietary intake on the postprandial glucose response while engaged in sustained versus interrupted sitting is unclear. This study aimed to the impact of uninterrupted sitting versus sitting with activity breaks on adolescents' postprandial glucose responses while consuming meals varying in energy (high-energy diet versus standard-energy diet).


Methods: Thirteen healthy participants (16.4±1.3 years) completed a four-treatment cross-over trial: 1) uninterrupted sitting + high-energy diet (meals consumed at 0 and 3h; consistent for each condition); 2) sitting with breaks + high-energy diet; 3) uninterrupted sitting + standard-energy diet; and 4) sitting with breaks + standard-energy diet. A Continuous Glucose Monitoring system (CGM) recorded interstitial glucose concentrations every five minutes. Linear mixed models examined differences in glucose positive incremental area under the curve (iAUC) and total AUC between the sitting and diet conditions for the first meal, second meal and entire trial period.

Results: Compared to the uninterrupted sitting conditions, the breaks condition elicited a 36.0 mmol/L/h (95%CI 6.6-65.5) and 35.9 mmol/L/h (95%CI 6.6-65.5) lower iAUC response after the first and second meal, respectively, but not for the entire trial period or for total AUC. Compared to the standard-energy diet, the high-energy diet elicited a 55.0 mmol/L/h (95%CI 25.8-84.2) and 75.7 mmol/L/h (95%CI 8.6-142.7) higher iAUC response after the first meal and entire trial, respectively. Similar response to the high-energy diet were observed for total AUC.


Conclusions: According to iAUC, interrupting sitting had a significant effect on lowering postprandial glucose for both dietary conditions, however, it was no longer significant when examining total AUC. Larger studies are needed to confirm these findings.

Clinical Trial Registration Number: ACTRN12615001145594.

Appendix F.4 Ethical approval



Human Research Ethics
Deakin Research Integrity



Memorandum

To: Prof Jo Salmon
School of Exercise and Nutrition Sciences
B
cc: Miss Elly Fletcher

From: Deakin University Human Research Ethics Committee (DUHREC)

Date: 02 April, 2015

Subject: 2015-039
The role of diet in the relationship between prolonged and interrupted sitting with postprandial glucose and insulin responses in adolescents
Please quote this project number in all future communications

The application for this project was considered at the DU-HREC meeting held on 16/3/2015.

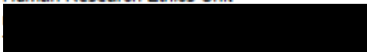
Approval has been given for Prof Jo Salmon, School of Exercise and Nutrition Sciences, to undertake this project from 2/04/2015 to 2/04/2019.

The approval given by the Deakin University Human Research Ethics Committee is given only for the project and for the period as stated in the approval. It is your responsibility to contact the Human Research Ethics Unit immediately should any of the following occur:

- Serious or unexpected adverse effects on the participants
- Any proposed changes in the protocol, including extensions of time.
- Any events which might affect the continuing ethical acceptability of the project.
- The project is discontinued before the expected date of completion.
- Modifications are requested by other HRECs.

In addition you will be required to report on the progress of your project at least once every year and at the conclusion of the project. Failure to report as required will result in suspension of your approval to proceed with the project.

DUHREC may need to audit this project as part of the requirements for monitoring set out in the National Statement on Ethical Conduct in Human Research (2007).

Human Research Ethics Unit


Appendix F.5 Parent Plain Language Statement and Consent Form



Date: Nov/Dec 2015
Full Project Title: Eat And Sit Youth (EASY) study
Principal Researcher: Alfred Deakin Professor Jo Salmon
Project Manager: Ely Fletcher



Plain Language Statement

To: Parents/guardian of adolescents aged 14-17 years,

Your child is invited to take part in the Eat And Sit Youth (EASY) research project.

This Plain Language Statement contains detailed information about the research project. Its purpose is to explain to you as openly and clearly as possible all the procedures involved in this project so that you can make a fully informed decision whether you are going to allow your child to participate.

Please read this Plain Language Statement carefully. Feel free to ask questions about any information in the document.

Once you understand what the project is about and if you agree to allow your child to take part in it, you will be asked to sign the Consent Form. By signing the Consent Form, you indicate that you understand the information and that you give your consent for your child to participate in the research project.

You will be given a copy of the Plain Language Statement and Consent Form to keep as a record.





Centre for Physical Activity and Nutrition Research



Purpose and Background of Research Project

The purpose of this project is to understand how different foods can affect how much sugar is in young people's blood when they sit for a long time and when they interrupt their sitting with light exercise breaks in between.

Up to 25 adolescents will participate in this project. Your child is invited to participate in this research project because he/she is aged 14 to 17 years.

What is involved in the research project

Participation in this project will involve your child attending Deakin University in Burwood on four separate days on weekends or during the school holidays. Each visit will be slightly different. On two of the research visits, your child will be asked to sit for 6 hours and eat two meals (one for breakfast, one for lunch). The other two research visits will be similar, but will involve your child getting up every 18 minutes and doing 2 minutes of light exercises (such as squats and step ups). During all four visits, your child may watch (age appropriate) television and movies that we have at Burwood, or play computer games of their choice. As a parent/guardian, you are most welcome to attend the research visits with your child, although it is not compulsory. Your child may also like to bring a friend if they would like some company during the research visits. If the friend is between 14-17 years, they are welcome to participate in the project along with your child.

See below for more details on what is involved in this research project for your child:

- 1. The day/night before each research visit:**

The day before all four research visits, we will kindly ask your child to not do any hard or tiring activities that will make them out of breath. This is because it may make your child tired during the research visits and it may influence the results. We will also give your child dinners for them to eat on each night before the four research visits. This dinner will be a familiar dinner that is commonly eaten by people of this age. This dinner is given to your child because we would like all participants to eat the same meal.

2. On the first research visit only:

- Your child's height, weight, waist and blood pressure will be measured privately by a research staff member. To measure your child's height, weight and waist, we will ask your child to remove any heavy clothing (e.g. jumpers and coat) and to take off their shoes. To measure their blood pressure, we will ask your child to sit quietly for five minutes before placing a cuff around their upper arm that inflates and measures their blood pressure. You may like to be there with your child for this, although again, not compulsory.
- Your child will be asked to complete two short surveys. The first survey is about your child's demographics, activity levels and how much time they spend sitting. Some questions that will be asked are: What is your date of birth? How much physical activity did you do last week? How much television do you watch each day? The second survey will ask your child to rate what stage they are at in their pubertal development. This involves your child circling a line-drawing that best describes their stage of puberty development. We will ask your child to complete this survey in private to avoid any feelings of discomfort. It is important for us to collect this information as the stage of puberty can affect a person's blood sugar levels. All survey responses are kept confidential.

3. On all four visits, your child will be asked to:

- Wear two monitors that record their activity: an accelerometer (pictured left) and an inclinometer (pictured right). The accelerometer tells us how much activity your child is doing and the inclinometer tells us how much time they are sitting. A staff member will fit the monitors to your child when they first arrive at Deakin, and remove them at the end of the day before your child goes home.
- Wear a monitor that records how much sugar is in your child's blood (pictured here). This monitor will be inserted into their lower back the night before they come to their first and third research visit. The monitor can be inserted either at Deakin University, or a research staff member can visit your child's home. We need to do this the night before as the monitor takes 12 hours to start working.



Page 3

The monitor can only be worn for 3-4 days, so after the second visit we will need to remove it and insert it again before the third visit. On the fourth research visit, we will remove the device again before your child goes home. Inserting this monitor does not hurt – we can show you and your child what it looks like and explain what it does prior to inserting it. Once the monitor is in, your child will barely notice it. We will adhere a plastic cover over the top so it won't move around. This monitor is water-proof so your child can still shower.

- Eat two meals during all four visits. Your child will be provided the first meal when they arrive at Deakin (approximately 8-9am) and the second meal 3 hours later (11am-12pm). The meals are everyday foods that are commonly eaten by people of this age. Your child will be asked to try and eat the whole meal within 20 minutes.
- Before and after each visit, we will ask your child to complete an 'appetite and mood questionnaire' to see whether sitting all day or eating different meals can affect their hunger levels and/or their mood (e.g. more alert or more tired).
- Throughout all four trials, we will ask your child to complete a short "physical activity and medication checklist". This will ask whether your child participated in any physical activity or taken any medications the 24 hours before the research visits.

To note: if you or your child has any concerns or questions about what's involved in the study or about the surveys or checklist, we are more than happy to answer any questions and/or show you all the study's materials.

4. At the end of the study:

As a thank-you for your child's time in this study, they will receive a summary of their results from the physical activity monitors and blood sugar monitor. They will also receive a FitBit Charge HR wristband tracker (picture right) on the last day of the study. The FitBit is a monitor that can record steps taken, energy expenditure, sleep patterns and has a built in heart rate monitor!





Centre for Physical Activity and Nutrition Research



Travelling to and from the research visits

If you would like to drive your child to and from Deakin University in Burwood, a parking permit will be available for you to use for the duration of the visit. If you are unable to drive your child, a myki card or taxi voucher will be provided to your child for them to use on their research visits to and from Deakin University. Prior to commencing the study, a research staff member will call you to discuss which travel option you would like your child to use.

Possible Benefits and Risks of Participating in the Project

This project will help us to understand how different foods can affect young people's blood sugar levels when they sit for a long time and when they sit with small breaks in between. We cannot guarantee or promise that participants will receive any direct benefits from this project. However, it is possible that by participating in the study, your child may become more aware of how much they sit during the day and what types of foods they consume. In addition to this, as a duty of care, if your child has any abnormal blood sugar levels, we will let you and your child know and recommend you see your local GP or Paediatrician.

We do not anticipate that there will be major risks involved in participation in this research project. However, when the blood sugar monitor is inserted, some participants may experience some discomfort where it is inserted. To reduce any potential discomfort, a topical anaesthetic cream (EMLA) may be applied to your child's skin as required. Also, the questions in Survey 2 (about the stage of pubertal development) may be sensitive or embarrassing to some participants. These questions are completed in private and are not shown to any other participants. However, if your child finds it uncomfortable, they do not have to complete it.

Privacy, Confidentiality and Disclosure of Information

All information collected will be used for research purposes only. Any information collected that can identify your child will remain confidential. To ensure confidentiality is kept, all project materials (e.g. survey, measurements collected) will be coded by a number and your child's name and personal details will be stored separately in locked filing cabinets at the School of Exercise and Nutrition Sciences, Deakin University.

Once the research project is complete, all hard copy research materials will be stored securely for a period of six years at Deakin University.



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Results of the project

Once the project has been completed and all data has been analysed, the results will be published in health, nutrition and physical activity journals along with both national and international conference presentations. In any publication or presentation, information will be provided in such a way that your child cannot be identified.

Monitoring of the Project

The lead researcher will monitor the progress of the research by having regular meetings with all research staff to discuss the progress of the project and address any issues that may arise during the research visits.

Funding

This research project is funded by a Centre of Research Excellence (CRE) Grant from the National Health Medical Research Centre (NHMRC). The research staff declare that there are no conflicts of interest.

Participation is Voluntary

Participation in any research project is voluntary. If you do not wish for your child to take part your child is not obliged to. If you decide to allow your child to take part and later change your mind, you are free to withdraw your child from the project at any stage.

Your decision whether to allow your child to take part or not to take part, or to allow your child to take part and then withdraw, will not affect your relationship or your child's relationship with Deakin University, or any other organisation involved.

Before you make your decision, a member of the research team will be available to answer any questions you have about the research project. You can ask for any information you want. Sign the Consent Form only after you have had a chance to ask your questions and have received satisfactory answers.

If you decide to withdraw your child from this project, please sign the Withdrawal of Consent form and post it back using the replied-paid envelope provided. Alternatively, you can notify a member of the research team on [REDACTED]



Centre for Physical Activity and Nutrition Research



Complaints

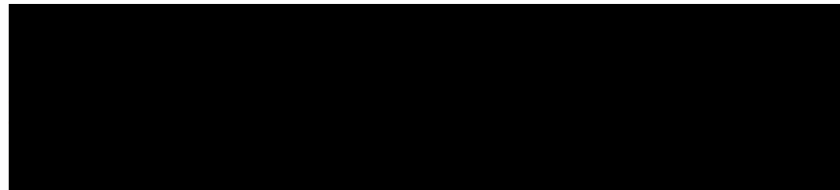
If you have any complaints about any aspect of the project, the way it is being conducted or any questions about your rights as a research participant, then you may contact:



Please quote project number: 2015-039

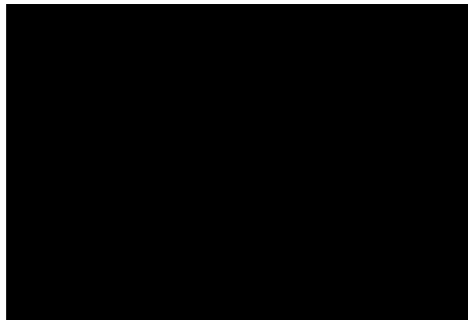
Further Information, Queries or Any Problems

If you require further information, wish to withdraw your child from the study or if you have any problems concerning this project, you can contact the Project Manager:



If you would like your child to take part in this study, please turn over the next page to complete the consent form. Please return the consent form to Deakin University in the envelope provided.

Thank-you for your time!





Centre for Physical Activity and Nutrition Research



ID:

CONSENT FORM

To: Parent/guardian of participant and participant

Date: 13/11/2015

Full Project Title: Eat Sit And Youth (EASY) study

Reference Number: 2015-039



I give permission for *(full name of your child)*
to participate in the project according to the conditions in the Plain Language Statement.

I acknowledge that:

- I have read and I understand the attached Plain Language Statement, and have been given a copy to keep.
- I agree for my child to participate in the assessment components of the project according to the conditions in the Plain Language Statement, and understand that parental/guardian consent is for my child to participate in the project.
- The researcher has agreed not to reveal my child's identity and personal details, including where information about this project is published, or presented in any public form.
- I am free to withdraw my child from this project at any time.

Parent/Guardian's Name (printed):
First name *Surname*

Home address:
Address *Suburb* *Postcode*

Contact number: (h) (m)



Parent/guardian's Signature (consent): Date:


Child's Signature (assent): Date:

Please return the signed consent form in the reply-paid envelope provided. If you have any questions, please contact the Project Manager [REDACTED]



Appendix F.6 Survey for female participants

ADMIN ONLY: EASY STUDY	
Participant ID:	
Today's date:	



EAT AND SIT YOUTH

EASY

Study

SURVEY - FEMALES

A: ABOUT YOU

A1. What is your birth date: _____ / _____ / _____
Day Month Year

A2. What year level are you in at school? Year _____

A3. What is your gender:

- ₁ Male
₂ Female

A4. In which group do you culturally identify yourself? *Note: you can tick one or more boxes*

- ₁ Australian
₂ Indigenous (including Aboriginal or Torres Strait Islander)
₃ UK
₄ New Zealand
₅ Indian
₆ Chinese
₇ Italian
₈ Vietnamese
₉ Greek
₁₀ Sri Lankan
₁₁ Filipino
₁₂ South African
₁₃ Other (please specify): _____

B: ABOUT YOUR PHYSICAL ACTIVITY

The following questions are about your physical activity.

'Physical activity' is any activity that increases your heart rate and makes you get out of breath at least some of the time. Physical activity can be done in sports, being active with friends or walking to school. Examples include running, brisk walking, cycling, dancing, skateboarding, swimming, soccer, basketball, football or surfing.

B1. Your usual physical activity levels

Over a typical or usual week, on how many days are you physically active for a total of at least 60 minutes per day? *(Please note, do not include school PE or school sport)*

- ₀ None
₁ 1 day
₂ 2 days
₃ 3 days
₄ 4 days
₅ 5 days
₆ 6 days
₇ 7 days (everyday)

B2. Vigorous activities

a) During the last 7 days, on how many days did you do VIGOROUS physical activities like heavy lifting, fast running or fast cycling?

_____ days per week

Tick this box if you did no vigorous physical activities, and SKIP to B3.

b) How much time did you usually spending doing VIGOROUS physical activities on one of those days?

_____ hours per day

_____ minutes per day

B3. Moderate activities

a) During the last 7 days, on how many days did you do MODERATE physical activities like carrying light loads, jogging at a light pace and cycling at a regular pace (do not include walking).

_____ days per week

Tick this box if you did no moderate physical activities, and SKIP to B4.

b) How much time did you usually spending doing MODERATE physical activities on one of those days?

_____ hours per day

_____ minutes per day

B4. Walking

a) During the last 7 days, on how many days did you WALK for at least 10 minutes at a time?

_____ days per week

Tick this box if you did no walking, and SKIP to Section C

b) How much time did you usually spend WALKING on one of those days?

_____ hours per day

_____ minutes per day

C: ABOUT YOUR SITTING TIME

The following questions are about your 'sedentary behaviours'.

"Sedentary behaviours" are things that generally involve sitting and not moving around, like watching TV, playing video games, reading, and playing on the computer.

Weekdays (Monday to Friday):

C1. Over a typical or usual week, on how many days do you use LESS THAN 2 hours of electronic media (e.g. watching television, seated electronic games and computer) for entertainment purposes each day?

- ₀ None
- ₁ 1 day
- ₂ 2 days
- ₃ 3 days
- ₄ 4 days
- ₅ 5 days
- ₆ 6 days
- ₇ 7 days (everyday)

C2. Please indicate how much time on a typical weekday (e.g., Monday to Friday) you do the following activities. Please think about the time from when you wake up until you go to bed.

Note: DO NOT include time when you are in school during regular hours, or weekends.

- a) Watching television/ videos/DVD's _____ hours and/or _____ minutes per day
- b) Playing sedentary computer or video games (e.g., Nintendo or Xbox) _____ hours and/or _____ minutes per day
- c) Using the internet, emailing or for leisure (e.g., checking Facebook) _____ hours and/or _____ minutes per day
- d) Doing homework (including reading, writing or using the computer) _____ hours and/or _____ minutes per day
- e) Reading a book or magazine NOT for school (including comic books) _____ hours and/or _____ minutes per day
- f) Riding in a car _____ hours and/or _____ minutes per day
- g) Riding in a bus, train, tram _____ hours and/or _____ minutes per day

Weekend (Saturday and Sunday):

C3. Please indicate how much time on a typical weekend day (Saturday or Sunday) you do the following activities. Please think about the time from when you wake up until you go to bed.

Please note: Do not include any weekdays.

- a) Watching television/ videos/DVD's _____ hours and/or _____ minutes per day
- b) Playing sedentary computer or video games (e.g., Nintendo or Xbox) _____ hours and/or _____ minutes per day
- c) Using the internet, emailing or for leisure (e.g., checking Facebook) _____ hours and/or _____ minutes per day
- d) Doing homework (including reading, writing or using the computer) _____ hours and/or _____ minutes per day
- e) Reading a book or magazine NOT for school (including comic books) _____ hours and/or _____ minutes per day
- f) Riding in a car _____ hours and/or _____ minutes per day
- g) Riding in a bus, train, tram _____ hours and/or _____ minutes per day

D: ABOUT YOUR PUBERTAL DEVELOPMENT

The following questions ask about changes that happen during puberty. Puberty is the time when your body develops into a young adult. As puberty can influence your blood glucose, the answers to these questions about your body are very important to us. Please do your best to answer carefully. If you do not understand a question or do not know the answer, please select the 'I don't know' option. Your answer will be kept private. Nobody can see your answers and we will not show them to anyone.

Breast development

1. Please circle the drawing that looks the most like your body.



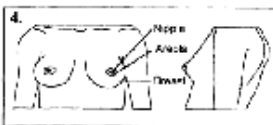
Stage 1: The breasts are flat. The nipples stick out a little.



Stage 2: The breasts are small mounds. The nipples stick out more than Stage 1.



Stage 3: The breasts and the darker skin around the nipples are bigger than Stage 2.



Stage 4: The nipple and the darker skin around the nipples make a mound that sticks out from the breast



Stage 5: Only the nipples stick out from the breast. The darker skin around the nipples does not stick out.

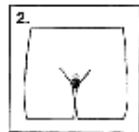
Please tick if you do not know

Female hair development

2. Please choose the drawing that looks the most like your body.



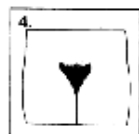
Stage 1: There is no hair in this area



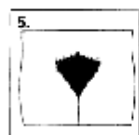
Stage 2: There are a few long, soft hairs in the private area. The hairs can be straight or curly.



Stage 3: The hair is thicker and curlier and has spread out over more of the private area than in stage 2



Stage 4: The hair is darker and curlier and covers a bigger area than Stage 3



Stage 5: There is hair on the inside of the thighs. The hair covers an area that is shaped like a triangle.

₆ Please tick if you do not know

Menstruation

3. Have you begun to menstruate (started to have your period)?

₀ No


₁ Yes.

If yes, at what age did you have your first period? _____ years

You are finished!
Thank-you for completing the survey 😊

Appendix F.7 Survey for male participants

ADMIN ONLY: EASY STUDY	
Participant ID:	<input type="text"/>
Today's date:	<input type="text"/>



EAT AND SIT YOUTH

EASY

Study

SURVEY - MALES

A: ABOUT YOU

A1. What is your birth date?: _____ / _____ / _____
Day Month Year

A2. What year level are you in at school? Year _____

A3. What is your sex?:

- ₁ Male
₂ Female

A4. In which group do you culturally identify yourself? *Note: you can tick one or more boxes*

- ₁ Australian
₂ Indigenous (including Aboriginal or Torres Strait Islander)
₃ UK
₄ New Zealand
₅ Indian
₆ Chinese
₇ Italian
₈ Vietnamese
₉ Greek
₁₀ Sri Lankan
₁₁ Filipino
₁₂ South African
₁₃ Other (please specify): _____

B: ABOUT YOUR PHYSICAL ACTIVITY

The following questions are about your physical activity.

'Physical activity' is any activity that increases your heart rate and makes you get out of breath at least some of the time. Physical activity can be done in sports, being active with friends or walking to school. Examples include running, brisk walking, cycling, dancing, skateboarding, swimming, soccer, basketball, football or surfing.

B1. Your usual physical activity levels

Over a typical or usual week, on how many days are you physically active for a total of at least 60 minutes per day? *(Please note, do not include school PE or school sport)*

- ₀ None
- ₁ 1 day
- ₂ 2 days
- ₃ 3 days
- ₄ 4 days
- ₅ 5 days
- ₆ 6 days
- ₇ 7 days (every day)

B2. Vigorous activities

a) During the last 7 days, on how many days did you do VIGOROUS physical activities like heavy lifting, fast running or fast cycling?

_____ days per week

Tick this box if you did no vigorous physical activities, and SKIP to B3.

b) How much time did you usually spending doing VIGOROUS physical activities on one of those days?

_____ hours per day

_____ minutes per day

B3. Moderate activities

a) During the last 7 days, on how many days did you do MODERATE physical activities like carrying light loads, jogging at a light pace and cycling at a regular pace (do not include walking).

_____ days per week

Tick this box if you did no moderate physical activities, and SKIP to B4.

b) How much time did you usually spending doing MODERATE physical activities on one of those days?

_____ hours per day

_____ minutes per day

B4. Walking

a) During the last 7 days, on how many days did you WALK for at least 10 minutes at a time?

_____ days per week

Tick this box if you did no walking, and SKIP to Section C

b) How much time did you usually spend WALKING on one of those days?

_____ hours per day

_____ minutes per day

C: ABOUT YOUR SITTING TIME

The following questions are about your “sedentary behaviours”.

“Sedentary behaviours” are things that generally involve sitting and not moving around, like watching TV, playing video games, reading, and playing on the computer.

Weekdays (Monday to Friday):

C1. Over a typical or usual week, on how many days do you use LESS THAN 2 hours of electronic media (e.g. watching television, seated electronic games and computer) for entertainment purposes each day?

- ₀ None
- ₁ 1 day
- ₂ 2 days
- ₃ 3 days
- ₄ 4 days
- ₅ 5 days
- ₆ 6 days
- ₇ 7 days (everyday)

C2. Please indicate how much time on a typical weekday (e.g., Monday to Friday) you do the following activities. Please think about the time from when you wake up until you go to bed.

Note: DO NOT include time when you are in school during regular hours, or weekends.

- | | |
|--|--|
| a) Watching television/ videos/DVD's | _____ hours and/or _____ minutes per day |
| b) Playing sedentary computer or video games (e.g., Nintendo or Xbox) | _____ hours and/or _____ minutes per day |
| c) Using the internet, emailing or for leisure (e.g., checking Facebook) | _____ hours and/or _____ minutes per day |
| d) Doing homework (including reading, writing or using the computer) | _____ hours and/or _____ minutes per day |
| e) Reading a book or magazine NOT for school (including comic books) | _____ hours and/or _____ minutes per day |
| f) Riding in a car | _____ hours and/or _____ minutes per day |
| g) Riding in a bus, train, tram | _____ hours and/or _____ minutes per day |

Weekend (Saturday and Sunday):

C3. Please indicate how much time on a typical weekend day (Saturday or Sunday) you do the following activities. Please think about the time from when you wake up until you go to bed.

Please note: Do not include any weekdays.

- a) Watching television/ videos/DVD's _____ hours and/or _____ minutes per day
- b) Playing sedentary computer or video games (e.g., Nintendo or Xbox) _____ hours and/or _____ minutes per day
- c) Using the internet, emailing or for leisure (e.g., checking Facebook) _____ hours and/or _____ minutes per day
- d) Doing homework (including reading, writing or using the computer) _____ hours and/or _____ minutes per day
- e) Reading a book or magazine NOT for school (including comic books) _____ hours and/or _____ minutes per day
- f) Riding in a car _____ hours and/or _____ minutes per day
- g) Riding in a bus, train, tram _____ hours and/or _____ minutes per day

D: ABOUT YOUR PUBERTAL DEVELOPMENT

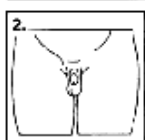
The following questions ask about changes that happen during puberty. Puberty is the time when your body develops into a young adult. As puberty can influence your blood glucose, the answers to these questions about your body are very important to us. Please do your best to answer carefully. If you do not understand a question or do not know the answer, please select the 'I don't know' option. Please note, your answer will be kept private. Nobody can see your answers and we will not show them to anyone.

Male penis development

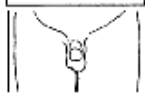
1. Please choose the drawing/description that looks the most like your body.



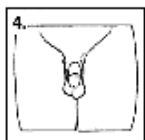
Stage 1: The penis, testicles, and scrotum are about the same size as when you were younger.



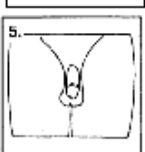
Stage 2: The testicles are larger than stage 1 and the scrotum is lower. The penis is only a little bigger compared to stage 1.



Stage 3: The penis is longer than Stage 2. The testicles and scrotum are larger and have dropped lower than in stage 2.



Stage 4: The penis is longer and wider than in Stage 3. The scrotum is bigger and the skin there is darker.



Stage 5: The penis, scrotum and testicles are bigger than in Stage 4

Please tick if you do not know

Male hair development

2. Please choose the drawing/description that looks the most like your body.



Stage 1: There is no hair in this area



Stage 2: There are a few long, soft hairs at the base of the penis. The hairs can be straight or curly.



Stage 3: The hair is thicker and curlier. There is hair growing on a bigger area than in Stage 2.



Stage 4: The hair is darker and curlier and covers a bigger area than Stage 3. There is no hair on the inside of the thighs.



Stage 5: The hair has spread to the inside of the thighs. The hair covers an area that is shaped like a triangle.

Please tick if you do not know

Deepening of voice

3. Have you noticed a deepening of your voice?

- ₁ Not yet started
- ₂ Barely started
- ₃ Definitely started
- ₄ Seems complete
- ₅ I don't know

You are finished!
Thank-you for completing the survey 😊

Appendix F.8 Participant 1-day food diary



The EASY study 1-DAY FOOD DIARY



PLEASE COMPLETE THIS ON:

If you have any questions,



HOW TO FILL IN THE 1-DAY FOOD DIARY

1. You will be asked to complete the 1-day food diary the day **BEFORE** all four research visits. That means, by the end of the study, you would have completed this four times.
2. Each page is divided into the main meals (breakfast, lunch and dinner) and snacks (morning, afternoon, and evening).
3. Beside each food and drink, please write a short **description** and the **brand** name, how it was **prepared** (e.g. fried, boiled, fresh), and the amount of food and drink you had (see example below).
4. Please try to record everything at the time of eating, NOT from memory at the end of the day. Otherwise you may forget some foods and drinks!
5. Many packet foods have weights printed on them, so please use these and give brand names whenever possible.
6. On the last page, please name any vitamins or other supplements you may have had that day.

EXAMPLE: HOW TO FILL IN A LUNCH MEAL		
FOOD/DRINK	DESCRIPTION & PREPARATION	AMOUNT
<i>Rolls</i>	<i>White</i>	<i>1 large</i>
<i>Butter</i>	<i>Devondale</i>	<i>Medium spread</i>
<i>Ham</i>	<i>Honey roasted, sliced</i>	<i>2 slices</i>
<i>Cheese</i>	<i>Tasty, full-fat</i>	<i>1 thick slice</i>
<i>Tomato</i>	<i>Fresh</i>	<i>3 slices</i>
<i>Water</i>	<i>Tap</i>	<i>600ml</i>
<i>Apple</i>	<i>Pink Lady, with peel</i>	<i>1 medium</i>

Breakfast

Time: _____

FOOD/DRINK	DESCRIPTION & PREPARATION	AMOUNT

Morning snacks

Time: _____

FOOD/DRINK	DESCRIPTION & PREPARATION	AMOUNT

Lunch

Time: _____

FOOD/DRINK	DESCRIPTION & PREPARATION	AMOUNT

Afternoon snacks

Time: _____

FOOD/DRINK	DESCRIPTION & PREPARATION	AMOUNT

Vitamins & Supplements

Please name any vitamins, mineral or other food supplements that you may have taken today. Please give all details and indicate what time you had them.

TIME	NAME & BRAND	AMOUNT
<i>EXAMPLE 8:00 AM</i>	<i>EXAMPLE Vitamin C, Blackmores</i>	<i>EXAMPLE 1 x 500mg tablet</i>

THANK-YOU FOR COMPLETING
THE 1-DAY FOOD DIARY



Please take this with you to your
next research visit

Appendix F.9 Participant checklist

ADMIN ONLY: EASY STUDY				
Participant ID:				
Today's date:				
Condition day #:	1	2	3	4
Condition type:	Sit-A ₁	Breaks-A ₂	Sit-B ₃	Breaks-B ₄

PARTICIPANT CHECKLIST

In addition to the 1-Day Food Diary, this checklist will ask you about the foods/drinks and medications you may have had yesterday and/or this morning, and whether you participated in any vigorous exercise or physical activity yesterday and/or this morning.

CHECKLIST 1: FOOD/DRINKS

The following questions ask about what you ate for dinner **LAST NIGHT**.

1. Did you eat the dinner last night that was provided by the EASY study? NO₀ / YES₁
2. Did you eat anything after 10:00 PM last night? NO₀ / YES₁
3. Did you drink anything after 10:00 PM last night? NO₀ / YES₁

The following questions ask about any foods and/or drinks you may have consumed **THIS MORNING** before your visit to Deakin University.

4. Did you eat anything this morning? NO₀ / YES₁
 If yes, what food(s) did you eat?: _____

5. Did you drink anything this morning? NO₀ / YES₁
 If yes, what drink(s) did you have: _____

CHECKLIST 2: YOUR PHYSICAL ACTIVITY

The following questions ask about any sport or exercise/activities that you may have done **YESTERDAY**.

1. Did you play or train for any sport team yesterday? (e.g. football, netball, rowing, tennis, swimming, soccer etc)

NO₀ / YES₁

If yes, what sporting activity did you do? _____

If yes, how many minutes did you play/train for? _____

2. Did you do any other exercise or activities yesterday that made you out of breath? (e.g. went for a run, went to the gym, played outside, etc)

NO₀ / YES₁

If yes, what exercise/activity did you do? _____

If yes, how many minutes did that exercise/activity go for? _____

The following questions ask about any sport or exercise/activities that you may have done **THIS MORNING**

3. Did you play or train for any sport team this morning? (e.g. football, netball, rowing, tennis, swimming, soccer etc)

NO₀ / YES₁

If yes, what sporting activity did you do? _____

If yes, how many minutes did you play/train for? _____

4. Did you do any other exercise or activities this morning that made you out of breath? (e.g. went for a run, went to the gym, played outside, etc)

NO₀ / YES₁

If yes, what exercise/activity did you do? _____

If yes, how many minutes did that exercise/activity go for? _____

Section G – Complete the morning of each research visit

2

CHECKLIST 3: YOUR MEDICATIONS

The following questions ask about any medications that you may have had **YESTERDAY**.

1. Did you take any **non-prescribed** medication yesterday? NO₀ / YES₁
 For example Panadol, Aspirin, Nurofen, etc.

If yes, what was the name of the medication? _____

If yes, how many did you take? _____

If yes, what was the reason for taking the medication? _____

2. Did you take any **prescribed** medication yesterday? NO₀ / YES₁
 For example, antibiotics, oral contraceptives, etc.

If yes, what was the name of the medication? _____

If yes, how many did you take? _____

If yes, what was the reason for taking the medication? _____

The following questions ask about any medications that you may have had **THIS MORNING**.

3. Did you take any non-prescribed medication this morning? NO₀ / YES₁
 For example Panadol, Aspirin, Nurofen, etc.

If yes, what was the name of the medication? _____

If yes, how many did you take? _____

If yes, what was the reason for taking the medication? _____

4. Did you take any prescribed medication this morning? NO₀ / YES₁
 For example antibiotics, oral contraceptives, etc.

If yes, what was the name of the medication? _____

If yes, how many did you take? _____

If yes, what was the reason for taking the medication? _____

THANK-YOU FOR COMPLETING THIS CHECKLIST

PLEASE RETURN IT TO THE RESEARCH STAFF

Appendix F.10 Food intake record (pre- and post-weight)

ADMIN ONLY: EASY STUDY				
Participant ID:				
Today's date:				
Condition day #:	1	2	3	4
Condition type:	Sit-A ₁	Breaks-A ₂	Sit-B ₃	Breaks-B ₄

Meal A: Breakfast

Start time: _____ End time: _____

Food	Pre-Weight (g)	Post-Weight (g)	Amount Consumed (g)
Bread (2 slices, 85g)			
Tomato (3 thin slices, 45g)			
Baby spinach (20g)			
Avocado (40g)			
Chicken, shaved (35g)			
Yoghurt (100g)			
Banana (120g)			
Apple (100g)			
Almonds (11)			
Water (350g)			
TOTAL			

Research staff initials: _____

ADMIN ONLY: EASY STUDY				
Participant ID:				
Today's date:				
Condition day #:	1	2	3	4
Condition type:	Sit-A ₁	Breaks-A ₂	Sit-B ₃	Breaks-B ₄

Meal B: Breakfast

Start time: _____ End time: _____

Food	Pre-Weight (g)	Post-Weight (g)	Amount Consumed (g)
Pizza (250g, 4 slices)			
Cheese (15g) *put on pizza			
Mars Bar (51g)			
Lemonade (350g)			
TOTAL			

Research staff initials: _____

ADMIN ONLY: EASY STUDY				
Participant ID:				
Meal eaten on (date):				
Before condition day #:	1	2	3	4
Before condition type:	Sit-A ₁	Breaks-A ₂	Sit-B ₃	Breaks-B ₄

Meal: DINNER # _____

Start time: _____ End time: _____

Food	Pre-Weight (g)	Post-Weight (g)	Amount Consumed (g)
Beef Lasagna (400g)			
Bread (1 slice, 30g)			
Mixed vegetables (50g)			
TOTAL			

* Note: Cross-check with 1-day food diary

Research staff initials: _____

Appendix F.11 Recruitment flyers and advertising

Example of flyer placed around Eastern suburbs of Melbourne.

The flyer features a teal header with the C-PAN logo (Centre for Physical Activity and Nutrition Research) and the Deakin University Australia logo. A blue speech bubble contains the text 'Attention Teenagers!'. Below this is a stick figure sitting on the ground. A list of three criteria is presented with checkmarks. A Fitbit Charge HR smartwatch is shown next to the text 'The EASY Study!'. A purple box states that participants will receive a Fitbit Charge HR as a thank-you. Contact information is provided in a teal footer, with some details redacted with black boxes.

C-PAN Centre for Physical Activity and Nutrition Research

DEAKIN
UNIVERSITY AUSTRALIA

Attention Teenagers!

- ✓ Are you aged between 14 and 17 years?
- ✓ Do you like watching TV and playing computer games?
- ✓ Are you available to attend Deakin University (Burwood) on 4 different days in January and February 2016?

If so, you may be able to participate in...

The EASY Study!

As a thank-you, all participants will receive a Fitbit Charge HR!

For more information, call [REDACTED] or email [REDACTED]

Centre for Physical Activity and Nutrition Research (C-PAN), School of Exercise and Nutrition Sciences
Deakin University Melbourne Burwood Campus 221 Burwood Highway, Burwood Vic 3125
[REDACTED]

Approved by Ethics DU-HREC #2015-039 Deakin University Cricos Provider Code: 00113B

Advertisement in a Melbourne local newspaper (“The Leader”), published 12 November 2015

ATTENTION PARENTS!

- Do you have a child aged 14-17 years?
- Do they like watching TV/movies & playing computer games?
- Are they always looking for something to do on weekends or during school holidays?

If yes, they might like to join...

The EASY Study

At the end of the study, your child will receive a Fitbit HR Charge!

For more information, please SMS your name and contact number to [REDACTED]

The EASY Study
Centre for Physical Activity and Nutrition Research (C-PAN)



Appendix F.12 Example of individualised report card given to participants

The **EASY** Study

Dear [REDACTED]

We would like to say **thank you** for your participation in the EASY Study. We hope you enjoyed the visits at Deakin University.

In this booklet, please see a summary of your blood sugar levels on all four visits:

Day 1: [REDACTED]

Day 2: [REDACTED]

Day 3: [REDACTED]

Day 4: [REDACTED]

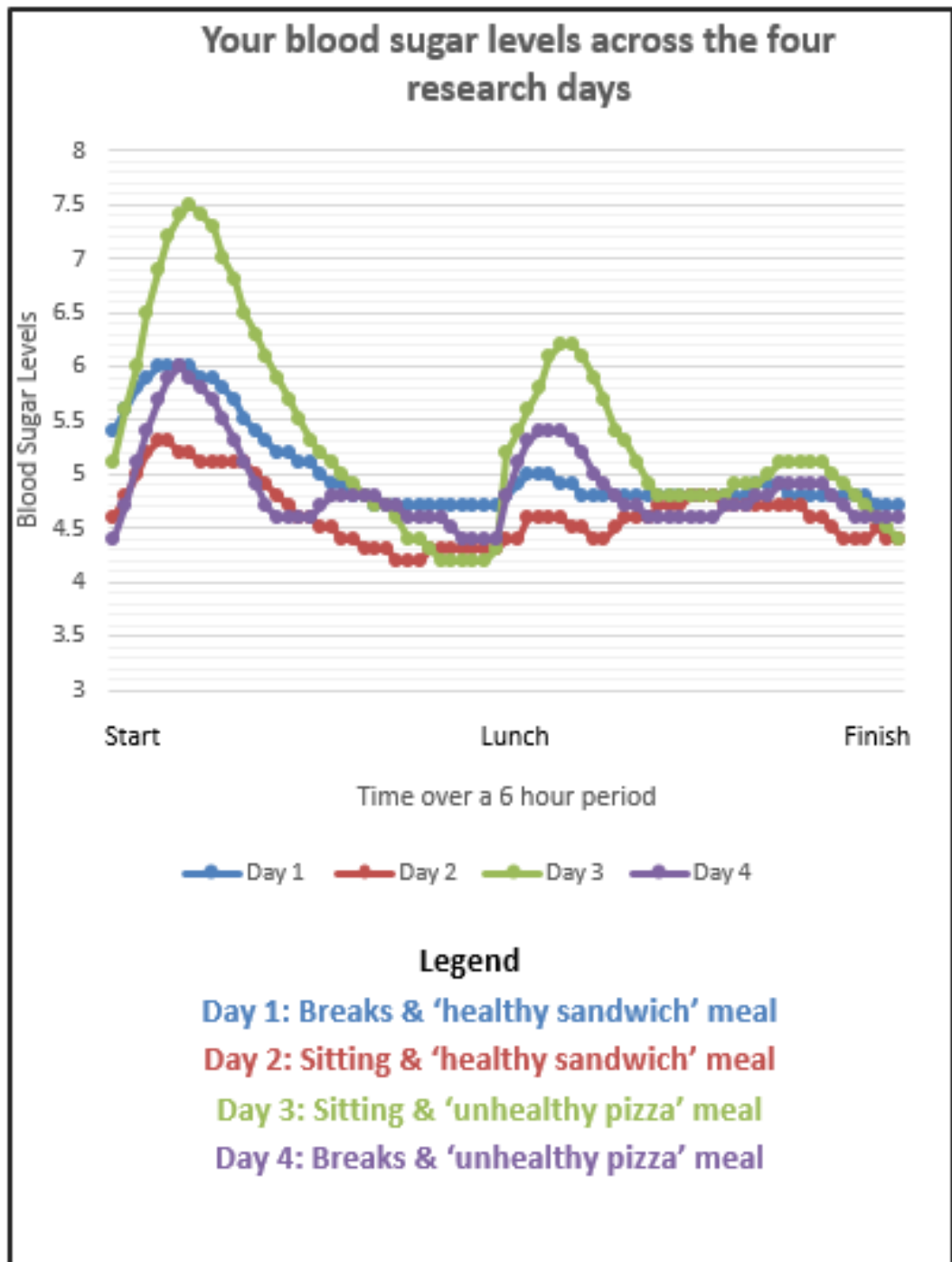
You will also find a summary of your height, weight, waist and blood pressure that were measured on your first visit, as well as an average of your fasting glucose measured over the 4 days.

Thanks again for participating in the EASY study!
Remember, to always try to break up your sitting time!

The EASY Research Team.



Your results...



Your measurements

Height: [REDACTED]

Weight: [REDACTED]

Waist circumference: [REDACTED]

Systolic Blood pressure*: [REDACTED]

Diastolic Blood Pressure*: [REDACTED]

Fasting glucose^: [REDACTED]

Recommendations for adolescents

*The healthy range for blood pressure in adolescents your age, is to have your systolic blood pressure less than 130 and your diastolic blood pressure less than 85.

^The healthy range of fasting blood glucose (sugar) in adolescents your age is between 3.8 to 8.3 mmol/L. The wide range is due to many factors influencing blood glucose during adolescence such as puberty, hormones, growth spurt, food and exercise.

Your Fitbit HR Charge!



We hope you are enjoying your new Fitbit HR Charge. Please see below some health recommendations to follow for adolescents your age. You can see whether you meet these recommendations on the Fitbit App.



Reach 10,000 Steps a day!



Do at least 60 minutes of 'active' physical activity each day!



Have 9-10 hours of sleep each night!



Drink 7-8 glasses of water each day!

Also, try to limit the time you spend using electronic media (e.g. television, laptop, iPad) for entertainment purposes to **less than 2 hours** per day.



Appendix F.13: Additional analyses - First peak glucose responses across four conditions

Table 1: First glucose peak responses across the four experimental conditions

	Experimental conditions				Effect (<i>P</i> -value)	
	High-energy diet + sitting	High-energy diet + breaks	Standard-energy diet + sitting	Standard-energy diet + breaks	Diet	Breaks
Glucose (mmol/L)						
First peak glucose	7.7 ± 0.3	7.0 ± 0.2	6.6 ± 0.3	6.1 ± 0.1	<0.001	<0.001
Difference from FG	2.7 ± 0.2	1.9 ± 0.2	1.8 ± 0.2	1.2 ± 0.2	<0.001	<0.001
Time (mins)						
Time-to-peak glucose [^]	35.3 ± 13.4	54.0 ± 13.3	38.3 ± 13.9	50.8 ± 13.5	0.992	0.275

Analyses comparing the diet conditions, and sitting/breaks conditions. Mean and SE estimated under a linear mixed model including period, diet, sitting and

% of meals consumed as fixed effects and participant as a random effect.

[^]Includes the allocated 20 minutes that participants had to consume the meal.

FG: fasting glucose

Appendix F.14: Additional analyses - Differences in percentage of meals consumed (high-energy versus standard-energy diets) between male and female participants

Table 2: Differences[^] in percentage of meals consumed (high-energy versus standard-energy diets) between male and female participants

	% high-energy diet Median (IQR range)	% standard-energy diet Median (IQR range)
Males	100 (98.4 – 100)	97.5 (76.2 – 100)
Females	76.5 (63.4 – 88.5)	87.7 (79.0 – 100)

[^] Differences between percentage of high-energy diet and standard-energy diet consumed across the entire trial compared between sexes using a Kruskal-Wallis test.

Significant difference observed between percentage of diets consumed by sex ($p = 0.019$).