ACUTE APPETITE, ENERGY INTAKE AND PHYSICAL ACTIVITY LEVELS OF 8 TO 11 YEAR-OLD BOYS IN RESPONSE TO ACTIVE VIDEO GAMING

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PhD Thesis

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ABSTRACT

There is evidence that physical activity (PA) levels are declining in English children. Sedentary screen based media activities, including computer use and video game play have been linked to low PA levels and unhealthy energy intake (EI). These behaviours appear to be particularly prevalent in boys during mid-to-late childhood. Recent laboratory-based studies have found active video games can increase children's energy expenditure (EE) and PA levels to moderate to vigorous levels, in comparison to matched conditions such as resting and seated video game play and so could increase children's PA. However, the previous active video gaming studies have utilised various protocols that probably do not accurately reflect the real-life active video gaming practices of children. Recently it has been established that there is EI in adolescents during active video gaming. If this is so, the EI during active video gaming could potentially counteract the EE from active video game play. The purpose of this thesis therefore, was to establish the acute appetite, EI and PA responses to active video gaming, in 8-11 y boys.

There is sparse information regarding the habitual active video gaming behaviours of children during mid-to-late childhood. Consequently, in study one a questionnaire was designed specifically, to understand the real-life active video gaming practices of 40, 7-11 y-olds. By utilising the survey findings, in study two an acute intervention was designed to investigate the subjective appetite sensations (hunger, prospective food consumption and fullness), EI and PA in response to active video gaming in 21, 8-11 y boys. Each boy completed four individual 90 min gaming bouts in a randomised order which were; 1) seated video gaming no food or drinks, 2) active video gaming no food or drinks, 3) seated video gaming, food and drinks offered *ad-libitum* and 4) active video gaming, food and drinks offered *ad-libitum*. The study determined that there were no differences in acute sensations of hunger, prospective food consumption and fullness, or EI (MJ) between the seated and active video gaming bouts during which foods and drinks were offered *ad-libitum* (bouts 3 and 4). Physical activity levels due to active video gaming were light

and from seated video gaming were sedentary. Energy intake during both bouts was greater than the estimated EE, thus producing a positive relative energy balance state in the boys.

As subjective appetite findings did not explain the high EI during both seated and active video gaming, the objective study of appetite was necessary. A previous adult study had established good reproducibility in GLP-$1_{7\text{-}36}$, glucagon, leptin and insulin by using the less invasive fingertip capillary sampling. Since fingertip capillary sampling had not been utilised to measure plasma concentrations of the above mentioned hormones in children during gaming, in study three, preliminary testing established good reliability for fasting plasma GLP-$1_{7\text{-}36}$ and blood glucose in 8-11 y boys. Enabling study three to compare acute satiety-related signalling, subjective appetite, EI and PA in 21, 8-11 y boys, in response to one bout of active video gaming and one bout of seated video gaming, during which food and drinks were offered *ad-libitum*. The satiety-related signals, namely plasma GLP-$1_{7\text{-}36}$ and blood glucose were measured alongside subjective appetite sensations and EI during active and seated video game play and in a post-gaming test meal. A significant increase in glucose showed the boys had consumed a greater proportion of carbohydrate (CHO) during active video gaming. However, as more total energy was consumed during seated video gaming, and plasma GLP-$1_{7\text{-}36}$ was higher during active video gaming, according to the 'glucostatic theory', there may have been a satiety response. The satiety signals may not have been strong enough to override the hedonic response to food intake, especially as fullness sensations were higher during active video gaming. Physical activity levels were light due to active video gaming and sedentary from seated video gaming and so on cessation of both bouts, the relative energy balance of the boys was positive. The positive relative energy balance state was then not compensated for by a reduction in EI in a post-gaming test meal. Instead, the additional EI resulted in an increase in the positive relative energy balance state, of the 8-11 y boys.

The overall findings of this thesis established that EI appears to be commonplace in the majority of 8-11 y children during active video gaming. Parents should encourage their children to play active, rather than seated video games to reduce sedentary time and also discourage EI during game play.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER V

LIST OF IMAGES

LIST OF ABBREVIATIONS

The following abbreviations have been used throughout the thesis and have been defined on first appearance in the text.

Abbreviation	**Meaning**
ANOVA	analysis of variance
AgRP	Agouti-related peptide
AUC	area under the curve
bpm	beats per minute
BBC	British Broadcasting Corporation
BMI	body mass index
BMR	basal metabolic rate
CCK	cholecystokinin
CHO	carbohydrate
CI	confidence interval
Co. Ltd	limited company
CV	coefficient of variation
CV%	percentage coefficient of variation
DLW	doubly labelled water
DPP-IV	dipeptidyl peptidase IV
DVD	digital versatile disc
EAR	estimated average requirements
EDTA	ethylenediamin tetra-acetic acid
EE	energy expenditure
EI	energy intake
$GLP\text{-}1_{7\text{-}36}$	active glucagon-like peptide 1
GSHR	growth-stimulating hormone receptor
HR	heart rate
HSE	Health Survey for England

kcal	kilocalorie
kJ	kilojoule
LOA	limits of agreement
METS	metabolic equivalents of time
MJ	mega joule
MVPA	moderate to vigorous physical activity
NPY	neuropeptide Y
P	participant
PA	physical activity
PAEE	physical activity energy expenditure
PAMS	physical activity measuring system
PC	personal computer
PRO	protein
PYY_{3-36}	peptide tyrosine tyrosine
REE	resting energy expenditure
SD	standard deviation
SEM	standard error mean
SPSS	Statistical package for social sciences
TM	trademark
VAS	visual analogue scales
vs	versus
$\dot{V}O_2$ max	maximal rate of oxygen consumption
WHO	World Health Organisation
©	corporation
®	registered

PREFACE

Peer reviewed publications that have arisen from this thesis

Allsop, S., Green, B.P., & Rumbold, P.L.S. (under review). The between-day variation in fasting satiety-related analytes in 8 to 11 year-old boys. *Physiology and Behavior*.

Allsop, S., Green, B.P., Dodd-Reynolds, C.J., Barry, G., & Rumbold, P.L.S. (2016). Comparison of short-term energy intake and appetite responses to active and seated video gaming, in 8-11 year-old boys. *British Journal of Nutrition*. doi: 10.1017/S0007114515005437.

Allsop, S., Dodd-Reynolds, C. J., Green, B. P., Debuse, D., & Rumbold, P. L. S. (2015). Acute effects of active gaming on *ad-libitum* energy intake and appetite sensations of 8-to-11 year-old boys. *British Journal of Nutrition*, 114(12), 2148-2155. doi: 10.1017/S0007114515003724.

Allsop, S., Rumbold, P. L. S., Debuse, D., & Dodd-Reynolds, C. J. (2013). Real-life active gaming practices of 7-11-year-old children. *Games for Health Journal: Research, Devleopment and Clinical Applications, 2*(6), 347-353. doi: 10.1089/g4h.2013.0050.

Conference proceedings that have arisen from this thesis

Allsop, S., Rumbold, P. L. S., Debuse, D., & Dodd-Reynolds, C. J. (2013). *A survey of the active gaming practices of 7-11 y-old children from the north east of England (UK).* Poster presentation at the International Society for Bahvioural Nutrition and Physical Activity, University of Ghent, Belgium.

Allsop, S., Rumbold, P. L. S., Green, B. P., Debuse, D., & Dodd-Reynolds, C. J. (2014). *Acute snack intake and appetite responses of 8-11 y old boys to active gaming.* Poster presentation at the Assoication for the Study of Obesity, UK conference on obesity, University of Birmingham, UK.

Invited presentation

Allsop, S., Rumbold, P. L. S., Debuse, D., & Dodd-Reynolds, C. J. (2013). *A survey of the active gaming practices of 7-11 y-old children from the north east of England (UK).* Oral presentation at the FUSE Physical Activity workshop, Newcastle University, UK.

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DECLARATION

I declare that the work contained in this thesis has not been submitted for any other award and that it is all my own work. I also confirm that the thesis has fully acknowledged the opinions, ideas and contributions of others.

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CHAPTER I

INTRODUCTION

1.1 Introduction

In England, the proportion of children meeting the recommended levels of PA is in decline. The Health Survey for England (HSE), an annual report of the nation's health behaviours, which includes data for children's PA and obesity levels, has evidenced this decline. In 2008, 28% of boys and 21% girls aged between 2 and 15 y met the advised levels of PA (Scholes & Mindell, 2012). The latest survey conducted in 2012, indicated that the proportion of children who were physically active at recommended levels, has decreased from 28% to 21% for boys and from 21% to 16% for girls. In North East England, the proportion of children who are physically active is even lower, with only 19% of boys and 16% of girls meeting PA guidelines (Scholes & Mindell, 2012). Being physically active is fundamental to the maintenance of good health and quality of life and is linked to obesity prevalence (Scholes & Mindell, 2012).

Childhood obesity has been increasing steadily in England and peaked in 2004, but there is still an upward trend and the most recent data defines 19.1% of children as obese (Niblett, 2014). In Newcastle upon Tyne (North East England), childhood obesity prevalence is higher with 23% of the City's children classified as obese (Public Health England, 2015). Furthermore obesity appears to have greater prevalence in boys during mid-to-late childhood (7-11 y) (Hughes, Sherriff, Lawlor, Ness, & Reilly, 2011).

Lack of PA has been linked to obesity (Scholes & Mindell, 2012) and both are associated with chronic non-communicable disorders related to the musculoskeletal and cardiovascular systems, type two diabetes and psychological illnesses (Varney, Brannan, & Aaltonen, 2014). Indeed, the decline in PA and increase in obesity in English children is beginning to lead to the aforementioned disorders, which are usually diagnosed in adulthood, becoming increasingly observed in children (Zimmet et al., 2007) illustrating, the need to address both sides of the energy balance equation [energy expenditure (EE) and EI].

At the same time as PA has declined, sedentary time for English children aged 2-15 y seems to have remained consistent (Eastwood, 2014). The most recent data shows that boys and girls are spending an average of 3.3 h and 3.2 h per day on weekdays being sedentary and this increases to 4.2 h and 4.0 h on weekends, for boys and girls, respectively (Eastwood, 2014). Coincidentally, children appear to be increasingly technologically driven, which could be a major contributor to their sedentary time. In the UK, 73% of children aged 5-16 y report having their own laptop, personal computer (PC) or tablet device and spend an average of 1.5 h online each day. They also spend an average of 2.5 h watching television every day (UK Youth, 2015). Furthermore, 100% per cent of 6-10 y children surveyed by the British Broadcasting Corporation (BBC), stated that they played seated computer games (Pratchett, 2005). Recently it has been suggested that during mid-to-late childhood (7-11 y), an increase in the aforementioned sedentary activities corresponds with a decrease in PA, and this appears to affect boys more than girls (Atkin, Corder, & van Sluijs, 2013).

Due to advancements in technology, children are increasingly spending more time sedentary and this unhealthy behaviour needs to change. In view of the accessibility children have to technology, the large amount of time they spend using it and the popularity of seated computer games, a potential solution to this is to combine exercise with video gaming. Active video games could therefore be used to increase PA and (or) reduce sedentary behaviour. Active video gaming requires movement to physically interact with on-screen images to play games that can replicate sports or dance. Paediatric research thus far has focussed on the effects of active video games on children's EE and PA levels. Findings have been positive and active video games have been shown to increase both EE and PA when compared with matched conditions such as resting, television viewing and seated video gaming (Graf, Pratt, Hester & Short, 2009; Lanningham-Foster et al., 2006; Maddison et al., 2007; White, Schofield & Kilding, 2011). However, the majority of these aforementioned studies have been conducted under laboratory conditions and have not provided rationale for the protocols used. The studies have utilised various active video gaming consoles and active video games and the duration of bouts has ranged from 5 to 30 min. The protocols therefore might not have resembled children's real-life active video gaming activity especially, in view of the timing of game play reported by adolescents in Canada (O'Loughlin, Dugas, Sabiston, & O'Loughlin, 2012) and The Netherlands (Simons, Bernaards, & Slinger, 2012) as being 50.5 min and 80 min per bout, respectively. Furthermore, active game play might also be affected by socio-demographic characteristics such as age and gender, since younger adolescents in The Netherlands were found to play active games more frequently than those who were older (Simons, Bernaards, et al., 2012), whilst in Canada, females were more likely to play the games than males (O'Loughlin et al., 2012).

Very little is also known about the reasons for paediatric active gaming. It has been established thus far that In New Zealand, children reported that the content of some active games was why they played them (Dixon et al., 2010). In The Netherlands adolescents consider them as being a more social form of computer game than non-active games, as they enable game play against others (Simons, Bernaards, et al., 2012). It has also been found that when children play active games with other individuals it increases their motivation to play them and this was believed to have reduced the dropout rate in an active gaming intervention (Chinapaw, Jacobs, Vaessen, Titze & van Mechelen, 2008). Information such as this has not been explored in paediatric groups, in the UK. Future research should therefore establish the real-life active gaming practices of children, to enable the design of studies that are more representative of this behaviour.

In relation to sedentary behaviour and specifically seated computer gaming, as well as a reduction in PA, there is also evidence of unhealthy EI during game play (Berentzen et al., 2014; Borghese et al., 2014; Pearson & Biddle, 2011). Two intervention studies that have examined the effects of EI from unhealthy foods and drinks, following bouts of seated video gaming are conflicting. One hour post-seated video gaming, 15-19 y males consumed more compared to resting (Chaput et al., 2011), whilst following 30 min of seated video game play 9-14 y boys consumed less in a test meal (Branton et al., 2014). Both were in positive energy balance immediately following the trials

(Branton et al., 2014; Chaput et al., 2011) which suggests that seated video gaming could contribute to positive energy balance in paediatric groups.

More recently, qualitative findings have described that adolescents have reported eating snacks during active video gaming. Moreover the longer the adolescents spend playing active video games, the greater the EI (Simons, Chinapaw, Brug, Seidell, & de Vet, 2015). It would have been anticipated that the extra movement required to play active video games, would potentially provide a deficit through increased EE. In contrast to seated video gaming however, if there is food available during active video gaming in paediatric groups, it is possible, that the extra energy expended may counterbalance the EI during game play. However, as yet, acute EI has not been measured during active video gaming in paediatric groups and considering the proposed rise in obesity prevalence (Hughes et al., 2011), the reduction in PA and increase in sedentary activity in children during mid-to-late childhood (Hughes et al., 2011), 7-11 y-old children would benefit from such investigation.

To date, there have been two studies that have measured EI following, active video gaming bouts in adolescents (Chaput et al., 2011; Gribbon, McNeil, Jay, Tremblay, & Chaput, 2015). Both studies established that in healthy and obese 15-19 y males, there was no increase in EI from a test meal after the 1 h bouts of active video gaming, when compared with matched seated video gaming and resting conditions (Chaput et al., 2011; Gribbon et al., 2015). However, at the end of the trials, the EI from the post-gaming test meal, exceeded the energy expended by active video gaming (Chaput et al., 2015; Gribbon et al., 2015). Although there has been a small amount of work conducted with adolescents, in relation to EI and active gaming there is nothing with younger children. Furthermore, the EI of younger children has not been measured during or following active video gaming.

In an effort to explain the underlying mechanism for the EI following the 30 min and 1 h bouts of seated gaming and 1 h bouts of active video gaming, the aforementioned studies also examined subjective appetite (Branton et al., 2014; Chaput et al., 2015, 2011; Gribbon et al., 2015). Sensations of hunger, prospective food consumption and fullness were measured using visual analogue scales (VAS). No differences were found in any of the aforementioned appetite sensations between seated video gaming and resting, in 15-19 y males (Chaput et al., 2011) or in 9-14 y boys (Branton et al., 2014). There were also no significant differences in subjective appetite sensations of hunger, satiety, prospective food consumption or fullness, between active and seated video gaming in the healthy (Gribbon et al., 2015) or obese 15-19 y males (Chaput et al., 2015). The lack of difference in appetite sensations between the gaming and resting conditions did not therefore provide an explanation for the positive relative energy balance state, at the end of the trials. As the previous studies did not find any underlying mechanism in relation to subjective appetite to explain the EI following seated video gaming, it might be more pertinent to investigate this during intervention. Furthermore, up to now, subjective appetite has not been studied during active video gaming in children and so this warrants investigation.

At the time of writing, one study had measured blood derived-appetite parameters in response to gaming but the findings were ambiguous (Chaput et al., 2011). No studies to date have examined satiety-related peptides in children during active video gaming. A reason for the absence of objective measurement during gaming in children could be the lack of low-invasive blood sampling procedures that are as reliable and valid as the antecubital-venous method. Therefore, there is potential to examine appetite by the measurement of satiety-related peptides in children, alongside subjective measures of hunger, prospective food consumption and fullness. Only recently, fingertip capillary sampling has been found to be in good agreement with the antecubital-venous method, in adults (Green et al., 2014). However, first of all, fingertip capillary sampling of satiety-related peptides from children requires preliminary work, to establish their day-to-day reproducibility.

1.2 Thesis purpose and aims

The overall purpose of this thesis was to establish the acute EI and appetite responses to real-life active video gaming in boys aged 8-11 y. The individual aims are as follows;

- To develop a questionnaire to survey the real-life active video gaming practices of 7-11 y children in the North East of England, UK. Then utilise the survey findings to design an active video gaming intervention that is representative of these active video gaming practices in an appropriate target population.

- To examine whether there are differences in subjective appetite sensations of hunger, prospective food consumption and fullness and EI from foods and (or) drinks offered *ad-libitum* to 8-11 y boys, during acute bouts of active and seated video gaming.
- To examine the between-day reproducibility of homeostatic appetite signals related to satiety; GLP-$1_{7\text{-}36}$, leptin, glucagon, insulin and glucose, in 8-11 y boys.
- To determine whether there are differences in EI, subjective appetite and homoeostatic appetite signals related to satiety found to have good day to day reproducibility, in 8-11 y boys, during and following acute bouts of active and seated video gaming.

CHAPTER II

LITERATURE REVIEW

2.1 Prevalence of paediatric obesity and overweight

Childhood overweight and obesity is described by the World Health Organisation (WHO) as one of the greatest health challenges in the world today (Sacks, Swinburn, & Xuereb, 2012). The life-limiting, physiological problems such as type two diabetes and cardiovascular diseases, as well as psychological disorders that can be caused by obesity can be tracked from childhood into adulthood (Niblett, 2015). Preventative measures therefore must be a high priority both in England and globally (Sacks et al., 2012).

Obesity and overweight in English children are defined using the UK National Body Mass Index (BMI) Reference Centiles, 1990 (Cole, Freeman, & Preece, 1995). The index designates the 85th to 90th centiles as childhood overweight and the 91st centile and above as obese (Cole et al., 1995). In England, an upward trend in overweight and obesity has been observed in 2-14 y children from 1995 (26%), which peaked to 33% in 2004. Recent findings from the Avon Longitudinal Study of Parents and Children quantified the peak incidence of overweight and obesity through childhood and adolescence within the study cohort. It established that obesity prevalence seems to peak in mid-to-late childhood, when aged between 7 and 11 y (Hughes et al., 2011).

Since 2004, childhood obesity trends have fluctuated marginally. The Health Survey for England began in 1995 and is an annual assessment of the health of children. The most recent survey reported obesity had risen slightly from 2012 and now 16% of boys and 15% of girls are estimated to be obese (Boodhna, 2013). Prevalence is highest in boys who are from families within the lowest income quintile (Boodhna, 2013). Data from the National Child Measurement Programme has also revealed that incidence of obesity in year six children (10-11 y) in England was 19.1% (Niblett, 2014). On a regional level, incidence is even greater in children from the North East of England with 21.1%, of those in year six being classified as obese. Newcastle upon Tyne, is a major city within the North East of England and has a population of 292,200 people. Such high incidence of childhood overweight and obesity has been linked to socio-economic status and Newcastle upon Tyne has several areas with levels of high deprivation (Newcastle City Council, 2014). Based on these data, it seems pertinent that obesity prevention strategies should be targeted in mid-to-late childhood in Newcastle upon Tyne. In doing so the risk of young people entering adolescence in a positive energy balance state could be reduced (Hughes et al., 2011).

2.2 Physical activity in children

In this section it is important to note that PA is measured in metabolic equivalents of time (METS) and this is used to describe the intensity of activity. The following MET thresholds are recommended for use with children, sedentary <1.5 METs; light 1.5 to <4 METs; moderate 4 to <6 METs; vigorous >6 METS (Trost, Loprinzi, Moore, & Pfeiffer, 2010). Physical activity in childhood is paramount in order to lessen the risk of chronic and life-limiting conditions that can occur at this specific life stage, or later on in adolescence and adulthood (Almond et al., 2011). Currently, overweight and obesity are a major challenge to child health and research has linked PA levels to weight status. Recent systematic reviews of published literature have revealed that, higher than normal body mass in children or increased EI, or indeed a combination of both are associated with low levels of habitual PA (Hills, Andersen, & Byrne, 2011). There also appears to be a relationship between high levels of PA (EE) and healthy body mass in children.

Low levels of PA or physical inactivity not only appears to increase the risk of overweight and obesity in children but there is increasing evidence which links this behaviour to various conditions that are detrimental to health, such as cardiovascular disease (CVD) (Andersen et al., 2006; Andersen, Riddoch, Kriemler & Hills, 2011), psychological health problems, musculoskeletal disorders (Hills et al., 2011; Kelly et al., 2013) and conditions related to the metabolic syndrome (Brambilla, Pozzobon, & Pietrobelli, 2011). Furthermore, the main therapeutic method that has been shown to improve health in relation to the aforementioned conditions, particularly obesity and musculoskeletal disorders, appears to be PA (Andersen et al., 2006; 2011; Brambilla et al., 2011; Janssen & LeBlanc, 2010).

In view of the negative health consequences of physical inactivity, the most recent PA guidelines for the UK have evolved from previous recommendations. In 2004, "At least five a week: evidence on the impact of PA and its relationship to health" (Bull et al., 2010) stated that children should be active at moderate to vigorous levels for at least 60 min per day. In addition, they should incorporate activities that strengthen, bone and muscle and improve flexibility (Bull et al., 2010). The guidelines outlined in "Start Active, Stay Active", now include recommendations for time spent being sedentary. They state that children aged from 5 to 18 y should engage MVPA for ≥60 min on a daily basis. There must also be participation in vigorous intensity activity that will strengthen bone and muscle, on at least 3 days per week. In addition, time spent being sedentary, which is defined as sitting or lying still for extended periods, should be kept to a minimum (Bull et al., 2010). However, the guidelines do not specify what is considered to be an extended period of time and so could prove confusing for parents and carers of children. In Canada and Australia, recommendations are more specific and advise that children should spend no more than 2 h per day watching television, playing seated electronic games and using computers (Australian Government. Department of Health, 2014; Tremblay et al., 2012).

Despite the greater detail provided in the UK recommendations, the most recent data indicates that children's PA levels have declined, with only 21% of boys and 16% of girls achieving moderate to vigorous activity for at least 60 min per day (Niblett, 2015), whilst in 2011, 32% of boys and 24% of girls aged from 5-15 y were reported to meet 60 min of MVPA per day (Eastwood, 2012). However, when PA was measured objectively in a sub-sample of children, it was revealed that only 21% of boys and 16% of girls were meeting PA guidelines (Health and Social Care Information Centre, 2011). The objective data thus suggests that the self-report (or parental report) data of children's PA is overestimated. Indeed the Gateshead Millennium Study, a longitudinal study in a cohort of North East children, compared parental reports of children's PA with accelerometer data and established that MVPA was overestimated by 122 min per day (Basterfield et al., 2008).

The European Youth Heart Study (2003) in which PA was measured objectively by accelerometry, a decline was observed that appeared to correspond with increasing age (Riddoch et al., 2004). There is also evidence of a decline in PA in English boys as they get older (Niblett, 2015; Scholes & Mindell, 2012). The Health Survey for England (2015) reported that when aged between 5 and 7 y, the proportion of boys meeting PA guidelines was 24% however, at 13 to 15 y only 14% were found to meet them (Niblett, 2015; Scholes & Mindell, 2012). Such a reduction in PA in boys entering adolescence, once again suggests that mid-to-late childhood (7-11 y) is a critical age at which PA interventions should focus. In doing so, there is greater likelihood that higher levels of PA will be maintained into adolescence and adulthood and a reduced risk of the associated ill health should follow (Riddoch et al., 2004).

2.3 Children's sedentary behaviour

Distinct from PA, physical inactivity and sedentary behaviours have become serious challenges for health (Basterfield et al., 2014; Blair, 2009). Indeed research suggests that children who are sedentary for prolonged periods of time are at increased risk of ill health and this appears to be independent of whether PA levels meet with recommendations (Tremblay et al., 2011). Sedentary behaviour is defined as sitting or lying down for extended periods (Bull et al., 2010). Physical inactivity is defined as being moderately active for <30 min on <5 days per week or vigorously active for <20 min on <3 days per week (Mendis, Puska, & Norrving, 2011). The latest Health Survey for England (Eastwood, 2014) estimated that time spent being sedentary for extended periods, exclusive of time at school, was 3.3 h for boys and 3.2 h for girls on weekdays. On a weekend, this increased to 4.2 h and 4.0 h for boys and girls, respectively (Eastwood, 2014).

It was previously thought that watching television was a major contributor to the increase in child physical inactivity, sedentary behaviour and obesity and so was used as the proxy measure for sedentary behaviour (Jakes, Day, Khaw, Luben, & et al., 2003). Findings of a systematic review of television viewing which examined published data from 90 studies conducted in 28 countries however, found this method was almost certainly unreliable. The review provided assessments of prevalence and age related trends in children up to the age of 18 y. It established that children between the ages of 7 and 12 y (mid-to-late childhood) spent an average of 105.5 min per day watching television. Boys spent more time than girls per day, with an average of 109 min and 102 min, respectively. Furthermore 29% of boys within this same age group spent more than 4 h per day watching television (Marshall, Gorely & Biddle, 2006). More recently, data from English children also found an association with socioeconomic status. As household income decreased, time spent watching television increased (Scholes & Mindell, 2012).

Although there is strong evidence that television is a major contributor to physical inactivity there is no proof that time spent watching television has increased over the preceding 50 y in children (Lake & Townshend, 2013). There must therefore be other contributors to the observed increase in child physical inactivity and sedentary behaviours. These could be a lack of green space in which to play outdoors safely (Lake & Townshend, 2013; Mitchell & Popham, 2008) and low access to sporting activities and equipment, due to cost (Scholes & Mindell, 2012). Moreover, when considering the ownership and usage of computers, tablets and seated video gaming consoles by children (Pratchett, 2005), sedentary screen based behaviours could also be major contributors. Systematic reviews of the effects of the aforesaid screen-based devices on children's sedentary behaviour have begun to reveal evidence of some detrimental health outcomes. Tremblay and colleagues (2011) reviewed the effects of sedentary behaviours including seated video gaming, non-school computer use and television viewing on body composition, fitness, risk factors for CVD and metabolic syndrome and psychological disorders of children and adolescents aged between 5-17 y (Tremblay et al., 2011). The findings were able to determine that exposure of > 2 h per day to the aforementioned sedentary pursuits, albeit primarily television viewing, led to greater BMI and waist circumference, decreased fitness levels and lower self-esteem (Tremblay et al., 2011). Chinapaw and colleagues (2011) also found moderate evidence of an inverse relationship between aerobic fitness in young people aged ≤ 18 y and screen time, yet in contrast to Tremblay et al., (2011), there were no associations with BMI (Chinapaw et al., 2011). Both of the aforementioned reviews were unable to determine any link between sedentary time and cardio-metabolic risk factors. More recently however, associations have been found between screen-based sedentary behaviours and obesity, insulin sensitivity, and cardio-metabolic risk (Mitchell & Byun, 2014). There is also evidence that suggests a weak association (Fröberg & Raustorp, 2014) between the sedentary time of 6-19 y children, measured objectively by accelerometry and increased glucose and insulin levels (Fröberg & Raustorp, 2014).

To date, the evidence of detrimental effects to health of sedentary pursuits is limited and a large number of studies have relied on less accurate self-report measures, instead of objective methods such as accelerometry (Chinapaw et al., 2011). Nonetheless, there appears to be sufficient support to suggest there might be an association between prolonged time spent undertaking sedentary behaviours and weight status in children (Saunders, Larouche, Colley, & Tremblay, 2012; Tremblay et al., 2011). In view of this, children should be encouraged to undertake pursuits which encourage greater movement, PA and less sitting (Fröberg & Raustorp, 2014; Tremblay et al., 2011).

When considering the evidence presented in this section, it is apparent that in English children, overweight and obesity is a problem particularly in mid-to-late childhood (7 to 11 y old), and is worse in boys (Boodhna, 2013; Hughes et al., 2011). From mid-to-late childhood, PA appears to decline, especially in boys and so interventions should be targeted at this specific population. Physical inactivity and sedentary behaviours are also increasing with no corresponding increase in television viewing, previously used as the proxy measure for sedentary behaviour (Jakes et al., 2003; Marshall et al., 2006). Furthermore, there is some evidence that more prolonged periods of time spent being sedentary are linked to increased weight status and decreased fitness, which could increase the risk of associated health problems.

2.4 Sedentary screen based media

Children are exposed to a wide range of screen based technology including television, electronic games, computers, computer games and the internet. The sedentary activity associated with these devices has been termed screen based behaviour (Huang, Wong, & Salmon, 2013). As the present thesis will explore active video gaming and the effects of this on EI and appetite, it is pertinent to also review the effects of screen based media upon children’s PA and food intake.

Research into screen based behaviour is problematic in terms of what should be measured and this has created dilemma as to whether certain activities should or should not be included. Consequently, many of the earlier studies focussed on television viewing and although this is a major contributor to sedentary behaviour, it is now not the only sedentary pastime. Activities such as reading, listening to music and homework, are increasingly being carried out by using screen technology (Huang et al., 2013). The measurement of children's sedentary behaviour therefore, should also include time spent in the various screen based media activities (Hands et al., 2011; Huang et al., 2013; Marshall et al., 2006).

Considering the wide range of screen based technology to which children are now exposed, it is possible that the use of these devices could be a causal factor for the decrease in PA levels (Niblett, 2015). Consequently, the effects of sedentary screen based behaviour on children's PA will be discussed. The Raine study (Hands et al., 2011) tracked children from 6 y until 14 y of age to examine the effects of screen time on PA and age. The study defined screen time as watching television and playing seated computer games. At 10 y, one third of both boys (31.0%) and girls (29.9%) were classified as being high screen users, which Hands and colleagues (2011) defined as spending ≥120 min per day using sedentary screen based technology. In addition, PA data established that 28.6% of the boys and 39.4% of the girls achieved only low levels. When later tracked at age 14 y, screen use was found to have increased with 52.8% of boys and 47.9% of girls classified as high screen users. Furthermore, the proportion of boys and girls established as having low PA levels had increased to 37.5%s and 53.0%, respectively. High screen use had thus become more prevalent particularly in boys, and low PA more widespread, the older they became (Hands et al., 2011). However, the measurement of screen time and PA was inconsistent and subjective, as the questionnaires that were utilised differed at each of the time points. Furthermore, PA was reported by parents at age 10 y and then self-reported by the children at 14 y and so both screen time and PA could have been underestimated (Aresu et al., 2009; Basterfield et al., 2008; Mindell, Coombs, & Stamatakis, 2014).

In Hong Kong, an even greater proportion (50.7%) of boys aged 10-12 y were found to spend more than 120 min per day using screen based media (Huang et al., 2013). The study measured both internet use and seated electronic gaming by questionnaire. The findings established that time spent using screen based media was higher in boys than girls (boys 62.4 min^{-1} versus girls 50.7 min^{-1}). In addition, the study explored potential correlates of both screen based media use and PA. In boys, family support for PA was negatively correlated with time spent using screen based media. Huang et al., (2013) suggested the greater screen use found in the Chinese boys might have been due to them being younger than the Australian boys and having greater reliance on parents for participation in structured PA (Huang et al., 2013).

Findings from the study of Sport, Physical Activity and Eating Behaviour: Environmental Determinants in Young People (SPEEDY), which measured time UK children spent using computers or a combination of sedentary screen based technology (Atkin et al., 2013), contradict those of Huang and colleagues (2013). The SPEEDY study also examined the presence of a computer in the bedroom and the sedentary time of the children at three separate time-points, at age 9-10 y, 10-11 y and at 13-14 y. The children's sedentary time was measured by accelerometry over a 7-day period, during waking hours. A minimum of 3 days of valid data comprising one weekend day and excluding time spent at school, was included in the measurement. Average screen time was subsequently found to increase with age in the children, particularly in boys, from 8.1 h per week to 15.2 h per week. Likewise, their sedentary time increased from 34.6 h per week at 9-10 y to 40.3 h per week at 13-14 y (Atkin et al., 2013). The greater independence with regards to screen based media from mid-to-late childhood thus led to an increase in screen time and a simultaneous decrease in PA, especially in the boys.

The accelerometer data collected in the SPEEDY study however, was only able to measure time spent being sedentary generally and was not able to distinguish actual screen time which was self-reported, as in the previous studies. Self-report data collection methods are known to be beset with inaccuracies and can be affected by recall and social desirability bias (Mindell et al., 2014). When compared with accelerometry data, it has been shown that parents report their children's PA, it is typically over-reported (Aresu et al., 2009; Basterfield et al., 2008). A further reason for the increase in sedentary time could be due to the GT1M being the accelerometer device utilised in the study. This particular model is only able to measure movement on one single plane and so accelerometer counts may therefore have been missed especially, as they were being used to measure movement associated with sedentary behaviour. Therefore, screen time might have been underestimated in all of the cited studies (Atkin et al., 2013; Hands et al., 2011; Huang et al., 2013) and so could PA could be lower than currently evidenced.

A similar study but on a much larger scale was conducted more recently with 5844 children aged 9-11 y, from Australia, Brazil, Canada, China, Colombia, Finland, India, Kenya, Portugal, South Africa, UK, and the USA (LeBlanc et al., 2015). The study found that on average the children were sedentary for 8.6 hours per day and 54.2% spent more than 2 h each day using sedentary screen based media. Furthermore, in boys those who reported greater use of sedentary screen based media were more likely to have higher BMI. In both boys and girls, the more time spent sedentary and using screen based media the less likely they were to meet PA guidelines and they were more likely to have high weight status and a television or a computer in their bedroom (LeBlanc et al., 2015). However, the study was still not without limitations as they generalised the findings and PA, sedentary time and sedentary screen based media would be expected to vary between countries due to environmental differences such as the weather.

2.4.1 Sedentary screen based media; child weight status and food intake

Considering the evidence provided in section 2.4 that during mid-to-late childhood, PA appears to decrease and overweight and obesity increase without a rise in television viewing, it is pertinent to discuss the potential effects of sedentary screen based media. In terms of weight status, a randomised controlled study provided initial evidence that time spent using screen based media affected child weight status (Epstein, Paluch, Gordy, & Dorn, 2000). Ninety obese 8-12 y old children were randomly assigned to one of four treatments, two treatments aimed to reduce television viewing and computer game play and another two to increase PA every day over either 10 or 20 weeks. The dose-response relationship was examined to establish whether weight reduction and increased fitness were attributable to reduced screen time or increased PA. Both were found to be effective, as they produced equal levels of weight loss and increased fitness in the children (Epstein et al., 2000). Screen time and PA however were measured by self-report and so whether the children's PA had in fact, improved was not evidenced as it was not corroborated by an objective measure, such as accelerometry. Furthermore, dietary intake was not measured and parents were aware that their children were participating in a weight-related study. As such there may also have been a change in the children's EI and so there is no tangible evidence that the weight loss was wholly due to the increase in PA or decrease in sedentary time.

Further evidence of a relationship between weight status and screen time was provided when both were measured in US children aged from 1-12 y (n=2831) (Vandewater, Shim, & Caplovitz, 2004). In the whole sample no association was found between television viewing and weight status. However in the children aged 9-12 y old, those with lower weight status typically used the computer for non-gaming purposes for moderate amounts of time. Those with higher weight status used it for non-gaming purposes either very little, or a lot. As this was only relevant to those aged 9-12 y, it was suggested that at this older age it was more likely that they used the computer for homework and email. The screen time data however was collected using 24 h time use diaries which were completed by a parent or carer (Vandewater et al., 2004). The reporting by some parents might also have been subject to social desirability bias, particularly those with children of high weight status (Aresu et al., 2009; Basterfield et al., 2008).

The Raine study (Hands et al., 2011) discussed previously in section 2.4, also investigated associations between screen time and weight status in boys and results were more consistent than those of Vandewater and colleagues (2004), possibly due to it being longitudinal. High screen use (watching television and computer game play for ≥120 min per day) increased from 22.2% at 8 y to 52.8% at 14 y. Along with this rise in prevalence of high screen use, overweight and obesity also increased from 15.8% at 8 y to 24.1% at age 14 y. However, as already mentioned (section 2.4.1), the data was collected by proxy parent report at 8 y and 10 y and then self-reported at 14 y.

In a more recent study that investigated the computer use and television viewing of 4072 children aged 4-13 y, the collection of dietary data was included (De Jong et al., 2013). Computer use was not found to be associated with being overweight. Yet in contrast to previous work, there was a relationship with television viewing for >1.5 h per day, but only with children aged 4-8 y. The children's diet was not found to be a causal factor for this relationship (De Jong et al., 2013).

However, there was low participation of children with low and middle socioeconomic status. Therefore, the study population was not representative of the city from which the data was collected.

In contrast to the findings of De Jong and colleagues (2013), a study of the non-advertising screen time and acute eating behaviour in children provided support for a dietary influence (Marsh, Ni Mhurchu, & Maddison, 2013). The study investigated the effects of television, video game play and recreational computer use, all of which did not include breaks for advertisements, on children's EI. There was evidence that television viewing led to overeating, but only a preliminary indication that screen based behaviours had this same effect (Marsh et al., 2013).

Further evidence of the detrimental effect of television on EI was provided when food intake, total sedentary behaviour and television viewing were measured in 9-11 y old Canadian children (Borghese et al., 2014). In agreement with previous findings of Pearson and Biddle (2011), a negative association was found between the frequency of consumption of healthy foods and television viewing, but not total sedentary time. There was also a positive association with the frequency of consumption of unhealthy foods and television viewing but not for total sedentary time. The researchers suggested that television viewing rather than total sedentary time was more indicative of these dietary patterns of behaviour. In this latter study however total sedentary time was measured by accelerometry which prevented the differentiation of time spent in individual screen based behaviours, including television viewing. The time spent watching television was measured by self-report frequency questionnaire, as was food intake (Borghese et al., 2014). As previously discussed, self-report methods inherently produce mis-reporting owing to recall and social desirability biases (Aresu et al., 2009; Basterfield et al., 2008; Livingstone & Robson, 2000).

More recent research has measured food intake, PA, weight status and screen time (television viewing and computer use) in children aged 11 y (Berentzen et al., 2014). The children whose screen time was $\geq$20 h per week were found to consume more portions of snacks per day (1.9 versus mean 1.3 portions) and were less PA (4.3 versus. 4.8 days per week). One extra hour of screen time per week was found to significantly increase BMI by 0.01 z-score and waist circumference by 0.11 cm providing evidence that greater screen time appears to lead to increased snacking, lower PA and higher weight status. A limitation of the study however, was that screen time was measured when the children were aged 11 y but the assessment of food intake, PA and weight status was one year later, when they aged 12 y (Berentzen et al., 2014).

The evidence reviewed in this section suggests that during mid-to-late childhood, high screen use becomes more prevalent and low PA more widespread which might stem from less parental influence on PA (Huang et al., 2013). Consequently, an increase in weight status could be due to either or both of these behaviours, collectively (Huang et al., 2013). The negative associations with healthy food consumption and positive associations with unhealthy food intakes during screen time (Berentzen et al., 2014; Borghese et al., 2014; Pearson & Biddle, 2011) add support for a detrimental effect of screen time on PA, EI and weight status, in mid-to-late childhood, especially in boys.

2.5 The phases of appetite; hunger, satiation, fullness and satiety

In view of the evidence described in section 2.4.1 which indicates a link between screen-based use and unhealthy EI and low PA, it would be appropriate to investigate appetite to provide clarification for these behaviours in children. Appetite encompasses four individual components or phases during food intake, namely hunger, satiation, fullness and satiety. The three phases of appetite have multidimensional facets which are sensory, cognitive and metabolic. Firstly, hunger denotes an individual's drive to eat which is associated with physical sensations such as, the stomach feeling empty, muscles feeling weaker or a light headedness (Gibbons & Blundell, 2015). The sensations are caused by the orexigenic hormone, ghrelin (Cummings & Overduin, 2007), the release of which stimulates the hormones, neuropeptide Y and agouti-related peptide that promote food intake (Wren et al., 2001) (discussed in greater detail in section 2.5.4). Further stimulants of food intake are caused by the integration of sensory and cognitive stimuli generated by olfactory sensations, the sight and sound of food and the anticipation of specific flavours (Yeomans, 2008).

As food intake progresses, the metabolic, sensory and cognitive sensations of hunger start to decrease and an individual begins to have sensations of fullness, during a phase known as satiation

(Mattes, Hollis, Hayes, & Stunkard, 2005). The satiation phase is the process that governs meal size and duration and begins the termination of a meal. Meal size and duration are determined by both habit and the oro-sensory experience of the food that is being consumed. More specifically, the main determinants of satiation are the amount of the meal an individual perceives they have eaten and how full they expect to feel once they have consumed it. Post-ingestive metabolic signals are believed to have little influence during satiation (Yeomans, 2010).

Satiety is a post-prandial phase that comprises a decrease in hunger, increased feelings of fullness and physiological processes that lead to the termination of food intake (Gibbons & Blundell, 2015). Fullness is felt during the satiety phase of a meal due to the distention of the stomach which is a physiological change caused by the presence of carbohydrates (CHO), proteins (PRO) fats and fibre, that instigate the release of the hormone, cholecystokinin (CCK) (Gibbons & Blundell, 2015). Within minutes of ingestion, hormones such as glucagon-like-peptide (GLP-1) and peptide tyrosine tyrosine ($PYY_{3\text{-}36}$) are released. Levels of these post-ingestive hormones are released slowly and can remain elevated for more than 3 h (Verdich et al., 2001). There is some evidence that cognitive processes relating to an individual's attention and memory are also involved in satiety. The memory of the most recent meal is thought to reduce the amount eaten however, this does not appear to have any effect on the release of hormones during the satiety phase (Yeomans, 2010.

2.5.1 Assessment of energy intake and appetite

When considering the influence that the different components of appetite (hunger, satiation and satiety) have on food intake and the evidence that ingestion during sedentary screen based media use, may increase body mass in boys during mid-to-late childhood, further research should utilise rigorous methods to assess EI. In doing so, it will provide a more accurate assessment of the amount of energy consumed during the use of screen based media and could be used to give insight into relative energy balance.

Assessment of children's EI is beset with challenges related to participant age, cognitive ability and weight status (Livingstone & Robson, 2000). Prior to the measurement of children's EI, the outcome of the assessment, the dietary component that requires measurement, the population group being investigated and the time period of monitoring, must all be considered (Bates, Nelson, & Ulijasek, 2005). Dietary assessment can be carried out using prospective and retrospective methods, all of which have strengths and limitations (Collins, Watson, & Burrows, 2010).

Retrospective approaches such as, diet history, 24 h recall and food frequency questionnaire are some of the most commonly utilised methods. These methods however are subject to bias caused by participant memory, which can then lead to underestimation of important elements such as portion sizes (Frobisher & Maxwell, 2003). Under-reporting of unhealthy foods and over-reporting of healthy foods have also been found to occur in children and when compared to total EI, measured by doubly-labelled water (DLW), both behaviours are known to increase with child age (Champagne, Bray, Kurtz, Monteiro, & et al., 2002).

Prospective methods of EI assessment include weighed food records and food checklists. Weighed food diaries are one of the most widely used measures which require the participant to weigh and record all food and drink items prior to and following ingestion. This particular method and indeed all of those previously mentioned, are only appropriate when an assessment of the previous EI of the individual is required over a designated period of time (Collins et al., 2010).

For short-term intervention studies, that require the presence of the researchers and the measurement of participant dietary intake in the interim, direct observation is the most appropriate method. In a controlled setting and when providing food and drinks *ad-libitum*, direct observation is a prospective method that gives the most accurate measure of EI. Prior to offering the food and drinks, they are weighed or measured so that each item consumed can be recorded. Anything left over is also weighed or measured, enabling the total amounts of food and drinks consumed to be calculated (Bates et al., 2005). The disadvantages of direct observation are that it is labour intensive and good literacy and numeracy skills are essential. The advantages of direct observation however outweigh the aforementioned disadvantages for short-term or acute intervention studies. Participants are not aware their food intake is being measured. Therefore, problems such as deliberate under-reporting, avoidance of certain foods to reduce total EI and unintentional under-reporting are usually averted (Macdiarmid & Blundell, 1998). Furthermore direct observation is a method that has been used by the more meticulous paediatric appetite studies that have monitored and assessed the food and drink intakes of participants during short-term interventions (Dodd, Welsman, & Armstrong, 2008; Rumbold, Dodd-Reynolds, & Stevenson, 2013; Rumbold, St Clair Gibson, Allsop, Stevenson, & Dodd-Reynolds, 2011; Rumbold et al., 2013).

It is considered rigorous to standardize EI prior to appetite intervention studies. Changes in volumes of EI and macronutrient composition affect the time of digestion and absorption (Claessens, van Baak, Monsheimer, & Saris, 2009), which subsequently influences appetite. It is desirable therefore for participants to be in the same post-ingestive state during EI interventions wherein appetite is also measured (Flint, 2000; Flint, Raben, Ersbøll, Holst, & Astrup, 2001). The method of standardization must however be suitable for the study population to ensure compliance and accuracy and prevent drop-out. Weighed food diaries are one of the most widely used methods of EI assessment and their accuracy has been corroborated with DLW in children aged 7 y and 9 y (Livingstone et al., 1992). At 9 y, the children with the help of parents were also noted to show enthusiasm and interest in the weighing and reporting their food intake (Livingstone et al., 1992; Livingstone & Robson, 2000). However, essential dietary data can still be inaccurately reported such as portion sizes and whether any food or drink item was left over, so the use of two separate methods combined, is recommended in paediatric studies (Livingstone & Robson, 2000).

To counteract potential missing detail from children's weighed food diaries, Dodd and colleagues (2008) utilised the two pass 24 h recall method in combination (Ashley & Bovee, 2003). This latter approach is recommended for use in studies in which the design includes a free-living element (Livingstone & Robson, 2000) and has proved successful in acute appetite studies with 8-12 y females. The 24 h recall method is advantageous due to it being inexpensive, time efficient, not dependent upon literacy level and it accumulates precise detail, thus has low participant burden (Hill, Rogers, & Blundell, 1995; McPherson, Hoelscher, Alexander, Scanlon, & Serdula, 2000) At the time Dodd and colleagues (2008) utilised the combined method of weighed food diary and 24 h recall it had not been assessed in terms of accuracy in a paediatric population. Such assessment came later, when the precision of the combined method was tested in the laboratory by Rumbold and colleagues (2011). The 24 h EI of 13-15 y old girls was monitored and measured by trained researchers and assessed against each participant's ability to accurately complete a weighed-food diary and then recall their EI, 24 h later. Even though the population in the cited study were of an age whereby underreporting can increase (Bratteby, Sandhagen, Fan, Enghardt, & Samuelson, 1998; Livingstone et al., 1992), there was good agreement for the method between the participants and researchers on a group level (Rumbold, et al., 2011). On considering the evidence from the above two cited studies, the combined method of self-reported weighed food diary and 24 h recall appears to be the most robust and efficient technique to ensure standardisation of food intake, prior to an appetite-related intervention in paediatric populations which includes a naturalistic element.

2.5.2 Subjective measurement of appetite using visual analogue scales

Subjective appetite or appetite sensations are typically measured using visual analogue scales in adults and children. They were developed from VAS commonly used to gauge pain, mood nausea, fatigue and dyspnoea so that the individual could rate how they feel prospectively at a designated time-point, on a fixed-point scale (Gift, 1989; Hill & Blundell, 1982; Mattes, Hollis, Hayes, & Stunkard, 2005; Wewers & Lowe, 1990). In the measurement of subjective appetite, it is usual for the individual to place a vertical mark on a 100 mm horizontal line. Paediatric VAS generally includes three statements that denote the separate components of appetite, namely hunger, prospective food consumption and fullness. At each end of the 100 mm horizontal line are opposing statements enabling the participant to rate their appetite sensations accordingly. The following statements are typically used in paediatric appetite studies (Figure 2.1);

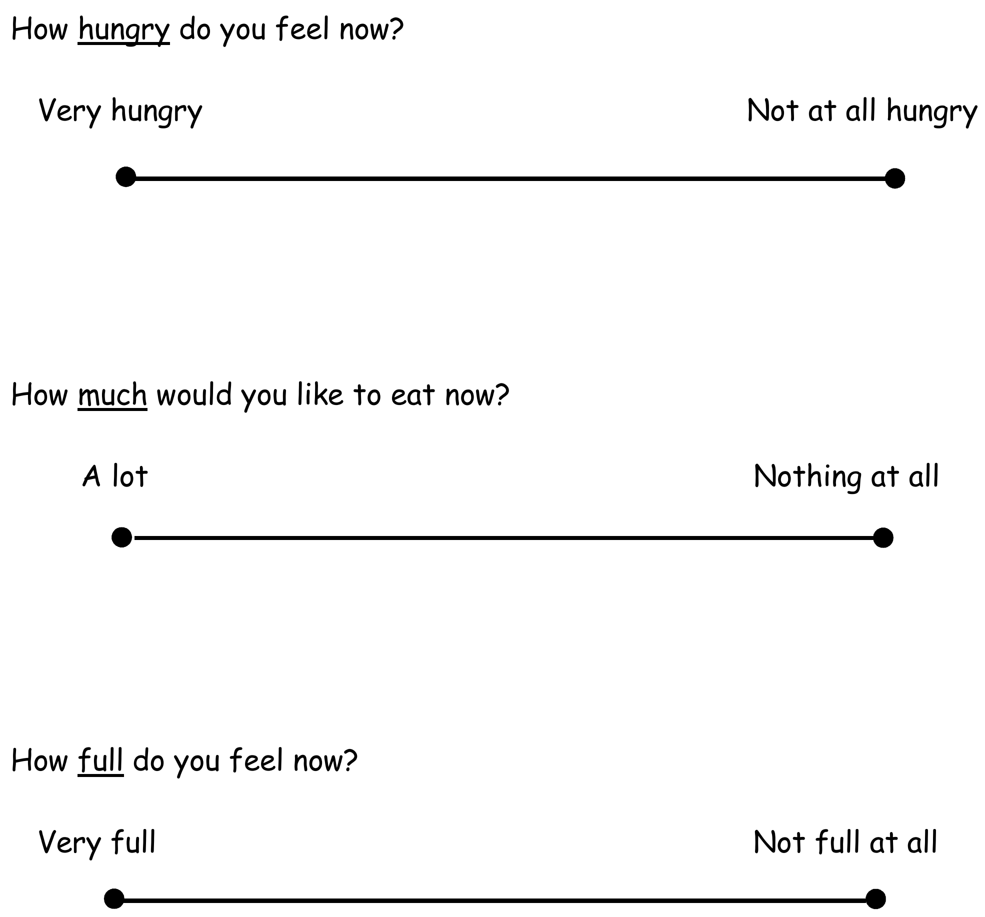

Figure 2.1. Typical visual analogue scales (VAS) utilised in the subjective measurement of appetite.

The use of VAS to measure subjective appetite has caused considerable debate in relation to the degree of understanding children have of rating systems. They have been found to have good reliability and validity in adult investigations particularly, when utilised in conjunction with another measure of eating behaviour in within-subjects studies with repeated measures (Stubbs et al., 2000). The scales have also been successfully implemented in previous paediatric appetite research with children aged from 8 y old and although a lot of data is believed to be ambiguous, there were no other suitable methods, at that time (Bellissimo, Thomas, Goode, & Anderson, 2007; Dodd et al., 2008; Rumbold, Dodd-Reynolds, et al., 2013; Rumbold et al., 2011; Rumbold et al., 2013).

2.5.3 Hedonic regulation of appetite

Human appetite is regulated by an intricate psychobiological system involving hormone signalling (biological), and cognitive and sensory stimuli (psychological). The regulation of appetite can be divided into three key phases, hunger, satiation and satiety. Hunger is instigated by the interaction of cognitive and sensory stimuli with the appetite hormone ghrelin. Satiety is brought about by biological changes in the gut which induce a cascade of complex hormonal signalling that act as a cue to end a meal and to stop eating. Satiety however, does not appear to be tightly biologically regulated. It seems that hormonal cues to end a meal can be overridden relatively easily by satiation, which is primarily a hedonic (or liking) response to foods that occurs before and during the ingestion of food.

Two key behaviours within hedonics which clarify how the foods ingested affect satiation, are palatability and food preference (Blundell & Finlayson, 2004; Yeomans, 1998). Palatability is an individual's perception that a particular food tastes good. Such a perception arises due to an interaction with the chemical structure of the food, their previous experience with it, nutritional needs at the time and the stage at which the food is tasted during a meal (Yeomans, 2008).

Food preference is the selection of one food over another, illustrated by when an individual is presented with more than one food at the same time. Food preference overlaps with palatability as food is preferred due to it being perceived as more palatable. Unlike palatability, preference does not have such a direct relationship with hedonics given that foods can be preferred for reasons such as, being considered healthier, more cost-effective or more nutritionally suitable (Yeomans, 2008). As there are no suitable paediatric studies, the importance of hedonics (liking, palatability and food preference), in relation to meal termination will be discussed from findings of an adult study. Participants were asked to eat a meal with varying levels of salt content which subsequently affected the palatability of it. The meal followed the consumption of either a low or high energy soup preload (Yeomans, 1998). Subjective hunger was increased with the low energy preload and was reduced from consumption of the high energy preload. However, during ingestion of the high energy preload followed by the more liked meal, the hunger ratings increased and eating time lasted 30 min longer. Whilst the high energy dense preload, in combination with the less palatable meal, caused hunger ratings to decrease at onset but changes during the meal were similar. Therefore, the energy density of the food has less of an effect on satiation, than liking (Yeomans, 1998).

2.5.4 Sensory evaluation of food

The hedonics of a particular food are coupled with the sensory evaluation of it. All individuals carry out a sensory evaluation of food before it is ingested. Sensory evaluation is stimulated primarily by the perception of flavour which derives from a complex assimilation of the senses. The senses involved are, gustatory which relate to the taste of the food, olfactory, being the smell of the food, also the temperature of it, the pain that may be caused from its ingestion e.g. chilli, the visual appearance and the sound it makes during and (or) after cooking (Zampini & Spence, 2004). Sensory evaluation is linked to survival and has a protective effect as it determines whether the individual accepts or rejects the food before putting it in their mouth (Yeomans, 2008).

The impact of sensory evaluation in relation to satiation was illustrated with adults by Wansink and colleagues (2005). In a laboratory controlled study, participants were asked to consume soup from a bowl that was either automatically and discretely refilled, or partly drained during eating. The volume of soup consumed was considerably more when being refilled (268 kcal) yet the meal was terminated earlier (157 kcal) when it was partly drained. In both trials, participants reported they had reached an identical level of satiety. The importance of which demonstrates that senses can override satiety-related hormones (Wansink, Painter, & North, 2005).

For the majority of individuals, eating is not only for survival but invariably, it is for pleasure. It is believed the anticipation of this pleasurable activity, might in itself be enough to stimulate hunger. The ‘liking’ for foods or ‘hedonics’ however is believed to exert considerable control over when an eating event starts and so determines how much we eat and when a meal is terminated (Yeomans, 1998, 2010). As demonstrated in children during television viewing (Temple, Giacomelli, Kent, Roemmich, & Epstein, 2007), there was a disruption in the physiological signals to terminate eating or what is referred to as a dishabituation to food cues (Epstein, Temple, Roemmich, & Bouton, 2009). In the previously cited study, normal weight 9-12 y children were randomised to a no television group, a television group that showed repeated clips of their favourite programme, or a group that was shown a continuous segment of their favourite programme. During all three trials the children were provided with a 1000 kcal portion of their favourite snack. At the beginning and end of the trials, hunger sensations were assessed using VAS. The trials lasted for 23 min or until the child indicated that they had consumed enough of the snack. Temple and colleagues (2007) established that the EI of children allocated to the continuous television group was significantly greater, they spent more time eating and there were no differences in hunger sensations between the three trials. Watching television continuously therefore disrupted the habituation to physiological food cues in this short period of time. Temple and colleagues (2007) suggested that the dishabituation to food cues had the potential to occur during computer and video game play. Both the cited paediatric study (Temple et al., 2007) and the study by Wansink and colleagues (2005) demonstrated that hedonics influence the physiological events in which peptides associated with satiation and satiety, are released post ingestion (Yeomans, 2008).

2.5.5 Biological regulation of appetite

Appetite-related peptides are secreted during two phases, to stimulate hunger and induce satiety. The release of the hormones relative to each phase, are controlled by the brain's hypothalamus and involve the actions and interactions of several peptides.

At present, acylated ghrelin is the only hormone known to be orexigenic and stimulate hunger during fasted periods or when the body is in a state of food deficit. Endocrine cells located in the stomach are the site of ghrelin's release into the circulating plasma. The presence of ghrelin, stimulates the release of neuropeptide Y (NPY) and agouti-related peptide (AgRP) which leads to the initiation of feeding (Cummings & Overduin, 2007; Wren et al., 2001). The identification of the ghrelin as an orexigenic hormone was relatively recent as it was initially believed to function purely as an endogenous ligand for the growth-hormone-stimulating receptor (GSHR) (Kojima, Hosoda, Date, Nakazato, & et al., 1999). The orexigenic actions of the hormone in humans were confirmed when it was shown that adult plasma concentrations increased during fasting and subsequently decreased following a meal (Cummings et al., 2001).

In response to food intake, the second biological phase is stimulated to induce satiety. There are several hormones, the release of which resembles a cascade of chemical signals. At the end of this there is satiety and thus termination of a meal. It is beyond the scope of the present work to discuss every hormone that contributes to satiety and eventual meal termination. Hormones such as CCK and $PYY_{3\text{-}36}$ are known to be released into the circulation from the I-cells and L-cells of the intestine respectively, and play central roles during the satiety phase (Batterham et al., 2002; Raybould, 2007). However, these hormones will not form part of the present study. A diagrammatic representation of the biological regulation of appetite is provided in Figure 2.2.

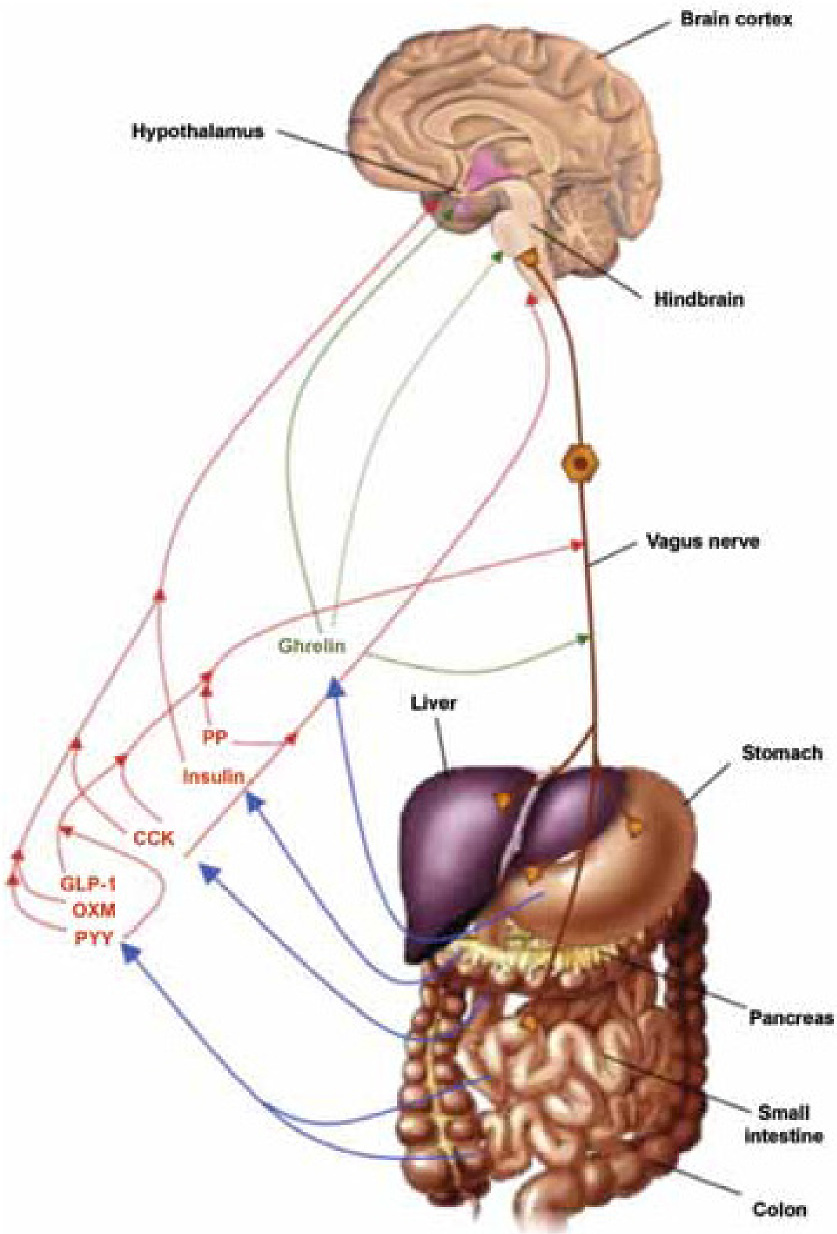

Figure 2.2. The biological regulation of appetite (Mendieta-Zerón et al., 2008).

2.5.6 Active Glucagon-like-peptide (GLP-$1_{7\text{-}36}$)

Glucagon-like-peptide (GLP-1), in the biologically active form GLP-$1_{7\text{-}36}$ is believed to have a central role in satiety and acts as a signal to terminate a meal. Active glucagon-like peptide 1, is released by endocrine L-cells located in the ileum only a few min following the ingestion of nutrients. The release of GLP-$1_{7\text{-}36}$ is stimulated by the presence of nutrients in the duodenum (Roberge & Brubaker, 1993). All macronutrients act as secretagogues however, GLP-$1_{7\text{-}36}$ is most sensitive to carbohydrates (CHO) (Ritzel, Fromme, Ottleben, Leonhardt, & Ramadori, 1997) and long-chain fatty acids (Roberge, Gronau, & Brubaker, 1996; Rocca & Brubaker, 1995). The actions of GLP-$1_{7\text{-}36}$ inhibit gastric emptying and intestinal motility, a process termed the 'ileal brake' (Williams, Baskin, & Schwartz, 2009). GLP-1 receptors are present in the gut, pancreas, brainstem, hypothalamus and vagal-afferent nerves. The biologically active form of the hormone is able to cross the blood-brain barrier although it is degraded very quickly by circulating dipeptidyl-dipeptidase IV (DPP-IV). As brainstem neurons have been found to produce GLP-1, it seems more likely the release from this region has more influence on the brain (Cummings & Overduin, 2007). Release from brainstem neurons facilitates action on sites thought to be located in the dorsal hindbrain (Kinzig, D'Alessio, & Seeley, 2002), from where hypothalamus regions involved in energy balance are activated to suppress eating (Tang-Christensen et al., 1996). In adults, levels of the circulating GLP-$1_{7\text{-}36}$ can be elevated for more than 3 h following a meal, so it not only acts to restrict food intake but also functions to extend the time before any further eating episode can occur (Verdich et al., 2001). In 9 y children, plasma concentrations of the peptide have been shown to elevate in response to high CHO and high fat meals served following a 30 min bout of light to moderate intensity exercise (Maffeis et al., 2012).

2.5.7 Glucagon, insulin and leptin

GLP-$1_{7\text{-}36}$ also aids in the regulation of blood glucose by inhibiting the release of glucagon (Drucker, Philippe, Mojsov, Chick, & Habener, 1987). Glucagon is secreted by α-cells located in the pancreas in response to falling concentrations of blood glucose and plasma insulin. The peptide interacts directly with the liver to maintain euglycaemia during fasted states and (or) when glucose is being utilised quickly and so is an antagonist to insulin (Jiang & Zhang, 2003). The interaction of glucagon with the liver during a meal indicates that in relation to satiety it is linked more to the control of meal size, rather than actual termination of eating (Geary, Le Sauter, & Noh, 1993 ; Woods, Lutz, Geary, & Langhans, 2006).

Glucagon is an antagonist to insulin which is released from pancreatic β-cells and coincides with the detection of macronutrients in the blood and so promotes glucose uptake thereby reducing blood glucose levels (Plum, Belgardt, & Brüning, 2006). As a result of this signal, there is a reduction in food intake (Woods et al., 2006) which is in accord with the 'glucostatic theory' in which insulin concentrations are augmented due to the increased presence of blood glucose (Mayer, 1952). Such a response is believed to indicate satiety.

Leptin is an adipokine largely produced by adipocytes and has long term pleiotropic and anorectic properties which inhibit EI and is correlated with white adipose tissue (Frühbeck, Jebb, & Prentice, 1998). When an individual is in a positive energy balance state, the leptin signal via capillary endothelial cells to the arcuate nucleus region of the medio-basal hypothalamus, is increased. The resultant effect of elevated leptin concentration in this region is increased sensitivity to other satiety-related signals which facilitates a reduction in food intake. Leptin signalling is long term as levels in the brain remain elevated until energy balance is restored (Woods et al., 2006). It should be noted however that there is much debate in relation to the effects of this peptide on satiety, as concentrations are higher in obese adults and children and is suggestive of leptin insensitivity (Guran et al., 2009; Halaas et al., 1997). Leptin also has specific short term functions that bring about a reduction of meal size. It appears to do this by acting on taste sensitivity through the hypopolarization of taste buds on the tongue (Kawai, Sugimoto, Nakashima, Miura, & Ninomiya, 2000) which reduces the positive reinforcing effects of food ingestion on the brain (Fulton, Woodside, & Shizgal, 2000). In obese children however, taste sensitivity appears to be become reduced (Overberg, Hummel, Krude, & Wiegand, 2012), illustrating there is potential leptin insensitivity.

2.5.8 *Objective measurement of appetite in children*

As yet, no paediatric gaming studies have measured satiety objectively during active video gaming by the quantification of plasma GLP-$1_{7\text{-}36}$, glucagon, insulin and leptin. Probable reasons for the lack of studies include the ethical challenges surrounding blood sampling from paediatric populations and the complexities associated with forward preparation of equipment, blood draw, storage and the specialist analysis of blood samples by assay (Cavender, Goff, Hollon, & Guzzetta, 2004). Rumbold and colleagues (2013) overcame these challenges and were the first to objectively measure hunger in response to exercise in adolescent girls. Before, during and following a bout of netball specific exercise versus resting, antecubital-venous blood samples were drawn to measure the responses of plasma acylated ghrelin, plasma insulin and blood glucose, in 13-15 y old girls. The reason for examination of plasma acylated ghrelin, was that several studies had observed an increase in hunger following exercise induced EE in children (Bellissimo, Thomas, Goode & Anderson, 2007; Bozinovski et al., 2009; Dodd et al., 2008; Rumbold et al., 2011). The study however observed no differences in plasma acylated ghrelin or plasma insulin concentrations following both the netball specific exercise and resting. Despite the lack of findings with regards to plasma acylated ghrelin, blood glucose levels were raised during the netball specific exercise. Furthermore, fullness sensations increased significantly 20 min into the netball specific exercise but not at the corresponding time-point during rest. Although the findings were not connected to the hunger peptide levels, the raised glucose levels were thought to be in accordance with Mayer's (1953) 'glucostatic theory' of appetite regulation (Rumbold et al., 2013). As mentioned in section 2.5.6, the 'glucostatic theory' proposes that an increase in blood glucose is indicative of satiety, thus hunger sensations in the girls could have been suppressed due to the netball specific exercise.

In relation to gaming, there has been only one seated video gaming intervention to have utilised both objective and subjective measurements of appetite. Chaput and colleagues (2011) compared the effects of a 1 h bout of seated video gaming versus resting, on EI, subjective appetite, total plasma ghrelin, serum insulin and blood glucose concentrations, in 15-17 y old males. Similar to Rumbold et al., (2013), there was no alteration in total plasma ghrelin, although once again a significant increase was observed in blood glucose levels due to the gaming condition which is indicative of satiety. Yet there were no differences in sensations of hunger, prospective food consumption or fullness and at the test meal following the two conditions, the EI of the participants was markedly greater after seated video gaming. Chaput and colleagues (2011) suggested that the participants were in a state of satiety due to seated video gaming and as such the EI response to seated video gaming was hedonic (Chaput et al., 2011). The proposed suppressive effects on hunger and suggested hedonic eating after seated video gaming, was speculative since there was no measurement of satiety-related hormones. The absence of a response in relation to hunger and ghrelin and the increase in blood glucose in response to the induced EE from seated video gaming, could have been more indicative of satiety. According to the 'glucostatic theory', glucose has a key role in short-term appetite regulation, as increased levels of blood glucose signify satiety (Mayer, 1952) thus increased concentrations of plasma satiety-related peptides (Woods, 2013).

If future paediatric appetite research does undertake the measurement of satiety-related peptides then the age and vulnerability of participants should be an important consideration. Recently the measurement of plasma GLP-$1_{7\text{-}36}$, glucagon, insulin and leptin concentrations by fingertip capillary sampling has been shown to agree with those drawn by the more invasive antecubital-venous method (Green, Gonzalez, Thomas, Stevenson, & Rumbold, 2014). Considering the associated interactions of the aforementioned satiety-related peptides with glucose, it is now possible to measure these in vulnerable populations. It should be noted however, that this is a new technique in the measurement of blood borne markers of appetite. Therefore, objective methods should be combined with subjective measurement in order to make comparison with previous work.

2.6 Active video gaming

Having discussed the relevant literature regarding sedentary screen based media, the following section will review the potential for active video games to increase children's PA or decrease sedentary behaviour. In 1987, the Foot Craz (Atari Inc.) (AtariAge, 2015) was the first active video game to be introduced (Image 1). The system comprised a small pad on which there were five coloured buttons that when pressed enabled a dance or running game. The Foot Craz (Atari Inc.) (AtariAge, 2015) was closely followed by The Power Pad in 1988 (Nintendo Co. Ltd, Kyoto, Japan) (Image 2) which was double-sided, larger and more complex. Both systems were limited in relation to the activities that could be played and were accessible in arcades only.

Image 1. An example of the Foot Craz (AtariAge, 2015).

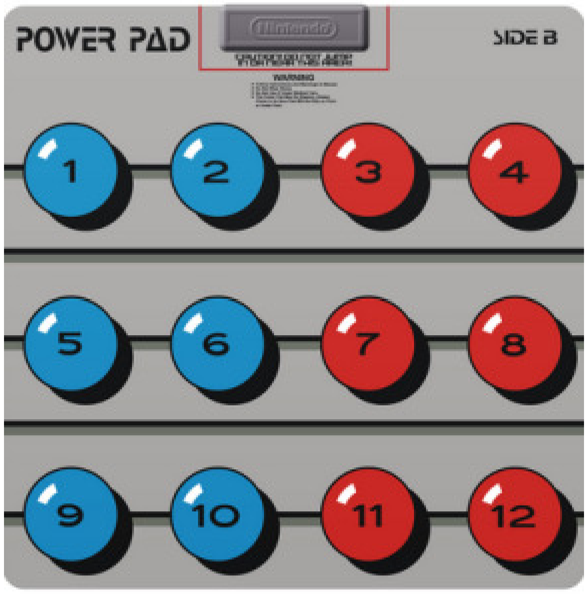

Image 2. An example of the Power Pad (Engadget UK, 2015).

The Eye Toy (Sony Computer Entertainment™) was then released in 2003 and was the first game to enable active video game play in the home. Linked to a television set, the console and games required the player to physically interact with on-screen images through a video camera that tracks their movements (Image 3).

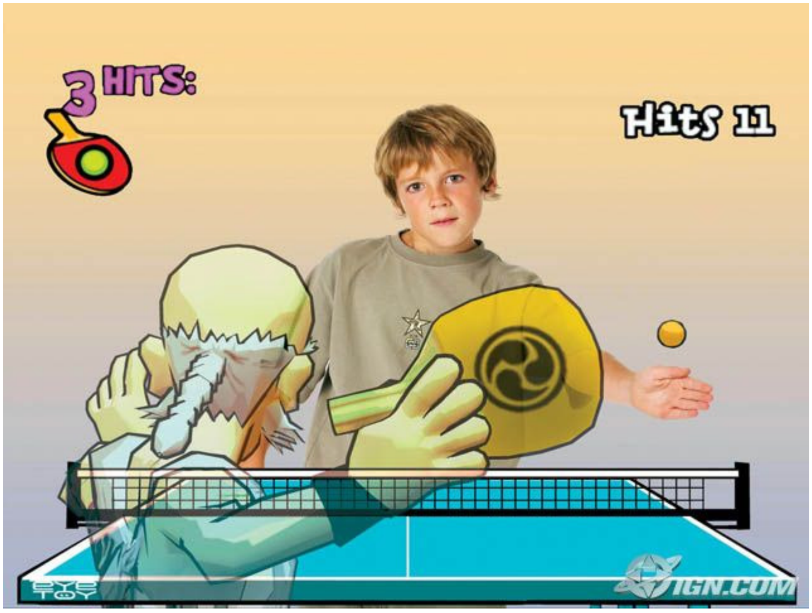

Image 3. An example of an Eye Toy game (Maddison et al., 2009, pp-149).

The Nintendo© Wii console was released only a few years after the Eye Toy (Sony Computer Entertainment®) in 2006. The Nintendo© Wii is the most recognisable system, having sold over 100 million consoles throughout the world (Johnson, 2013). The console allows games that replicate sports and dancing to be performed through a wireless handheld controller (Nintendo©, 2015) (Images 4 and 5).

Image 4. An example of people playing Nintendo© Wii Sports with the handheld controller (Reynolds, 2014).

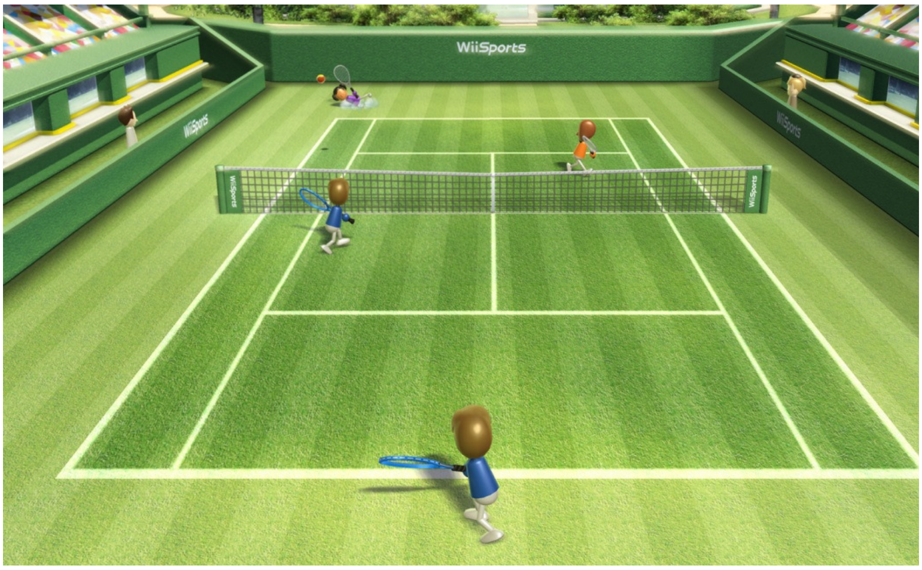

Image 5. An example of a Nintendo© Wii Sports game (tennis) (The Hut Group, 2015).

One year later (2007), Nintendo© introduced the Wii balance board, a peripheral that tracked an individual's movement and projected it onto the television screen and combined fitness with fun (Image 6). In 2010, the Xbox 360 Kinect (Microsoft©, 2015) was released which was the first hands-free active video gaming console (Image 7). The peripheral (motion controller) contains a web-cam device that enables the individual to interact with on-screen avatars (computer characters) without the need of a hand-held controller and so requires full body movement. Considering the popularity of active video games (Johnson, 2013) and the potential detrimental effects of sedentary screen based media to health (discussed in section 2.1), research began to explore whether active video gaming could feasibly be used to increase children's PA.

Image 6. An example of Nintendo© Wii Fit with the Wii balance board (GamingUnion.net, 2015).

Image 7. An example of hands-free game play, with the Xbox 360 Kinect (Microsoft©) (Privately Public, 2015).

2.6.1 Paediatric active video gaming behaviours

A survey by Simons and colleagues (2012) was the first descriptive study of active video gaming and was conducted in adolescents aged between 12-16 y. The survey was designed to make comparison between regular active gamers (≥1 h per week) and those who did not play active video games regularly (<1 h per week), in relation to socio-demographic characteristics, the time spent active video gaming and whether this replaced more or less active pursuits. The main findings revealed no significant differences in socio-demographic characteristics (age gender, ethnicity, education level of parent and child and sedentary behaviour) between the 201 adolescents who regularly played active video games and those who did not. More than half of the population (52%) were regular active gamers and on average, they then played for 80±136 min per week and this accounted for ~11% of their total PA. Furthermore, when active video gaming time was included in total PA time, the proportion of adolescents who met recommendations increased significantly from 67 to 73%. Lastly, they reported that their active video game play mostly replaced sedentary screen based behaviours (Simons, Bernaards, et al., 2012). Despite the useful contextual information gathered in the study, crucial data pertaining to the frequency of game play and type of active gaming consoles and games most frequently played by the Dutch adolescents' was missing.

Information was however obtained about the type, timing and intensity of active video gaming from 14-19 y old adolescents (n=1241) by O'Loughlin and colleagues (2012) and then later by Simons et al., (2013) in 373, 11-17 y olds. In comparison to the Dutch adolescents, only 24% of the Canadian adolescents reported that they played active video games. The reported game play was similar, as on average the Canadian adolescents played twice per week with each session lasting ~50 min. The Dutch population usually played active video games on 1.5 days during the week and on 1 day per weekend, for 36±32.9 min and 42± 36.5 min, respectively. The majority of Canadian adolescent active gamers played them at a moderate intensity and Nintendo® Wii Sports was found to be the most popular game (68%) (O'Loughlin et al., 2012). Information such as this enables active video gaming based interventions to be tailored to specific populations. However, the Canadian survey included questions related to lifestyle and body mass which may have induced social desirability bias. As such, the responses to the games played and the frequency and timing of play could have been over-reported.

Similar to the Canadian survey (O'Loughlin et al., 2012), an study into the prevalence of active video gaming conducted in 14-18 y US adolescents (n=9125) included the assessment of BMI (kg/m^2) (Song, Carroll, Lee, & Fulton, 2015). Findings of this US survey established that 40% of the adolescents participated in active video gaming at least $\geq$1 day per week. Furthermore, in this particular population, those who were active gamers tended to be the younger adolescents of black or non-Hispanic ethnicity. They also spent more time watching television or DVD's and were either overweight or obese but met PA guidelines. The inclusion of the BMI measurement however may have once again induced social desirability bias and resulted in over-reporting especially, in relation to meeting PA recommendations.

The most recent survey of (n=250) Dutch adolescents (mean age13.9±1.5 y), revealed some valuable personal and social factors related to their active video game play. Within the study population, 33.2% played active video games for $\geq$1 hr per week. Their game play was found to be associated with a positive attitude to the activity and playing active video games habitually, whilst they viewed non-active video games, negatively. Playing active video games with siblings or friends was also related to a sustained engagement in game play (Simons, de Vet, et al., 2014).

The cited surveys reveal valuable information regarding the active video gaming activity in adolescent populations in The Netherlands, Canada and the USA. The diverse findings particularly in relation to the differing prevalence of active video gaming and timing of game play, highlights the necessity for this type of study prior to PA and EE interventions. To date, this information is only available for adolescents and not for children. Data has not been collected from young people in the UK where physical inactivity is particularly problematic (Niblett, 2015).

2.6.2 Qualitative exploration of paediatric active video gaming

Similar to survey studies, there appears to have been only a handful of studies that have explored paediatric active video gaming qualitatively. The main purpose of the studies was to obtain specific information to inform future interventions. The first active video gaming focus groups were conducted with 37 New Zealand children, aged 10-14 y and their parent/s or main carer/s to explore facilitators and barriers to active video game play (Dixon et al., 2010). Predilection for active games appeared to be dependent upon age and gender, as girls and older boys favoured those that contained dance and music whilst the younger boys were motivated by aggressive action. The majority of children preferred the content of seated video games to active video games as these were considered more challenging although, along with the parents and main carers, they recognised that active video gaming could be used to increase PA and fitness (Dixon et al., 2010). Despite these optimistic findings, the views could not be generalized due to this being an initial study conducted in a small sample of children and parents.

Further focus groups with 37, 12-16 y-old adolescents then also established that content of active video games was considered to be less varied than seated video games by (Simons, de Vet, et al., 2012). However the adolescents reported that they mostly played active video games with friends or at parties and was a feature that they enjoyed about these games. Likewise, 46, 8-12 y-old children and their parents (n=19) viewed active video game play as being a more sociable form of gaming, as it enabled them to play against friends or family (De Vet, Simons, & Wesselman, 2014). Furthermore in contrast to the previous populations, they preferred them to seated video games and parents were happier to buy their children active video games since they required PA. Nonetheless parents did not perceive active video gaming as a replacement for outdoor play or sport (Simons, de Vet, et al., 2012).

The findings of the active video gaming surveys and focus groups thus far have provided valuable information needed to design and inform interventions. The information provides an indication of the prevalence of active video gaming, the time and frequency of game play and estimates of the PA levels they induce. Both the children and parents appear to hold positive perceptions of active video games particularly in relation to the physical aspect of them and the potential they have to increase PA and (or) reduce sedentary behaviours. There are some differences in findings between study populations', possibly due to the limited information that qualitative data can provide. As such future survey of children's active video gaming may benefit from both structured and open-ended questions.

2.6.3 Children's active video gaming energy expenditure

In 2006, research began to examine the EE produced from active video game play to determine whether the games could be utilised to increase children's PA. In 2006, Lanningham-Foster and colleagues (2006) were the first to measure active video gaming EE in a paediatric population. Energy expenditure was measured by indirect calorimetry in the laboratory in 8-12 y old children during two 15 min active video gaming bouts. Two active video games were utilised, Nicktoons Movie (Sony Eye Toy) and Dance Dance Revolution, Ultramix 2. The energy expended was compared with 15 min of EE in a rested state (REE), seated television viewing, seated video gaming and walking on a treadmill at 1.5 mph whilst watching television. The active video games produced the greatest EE in comparison to REE, with Nicktoons Movie (Sony Eye Toy) and Dance Dance Revolution, Ultramix 2 eliciting 273±101 $kJ \cdot h^{-1}$ and 382±181 $kJ \cdot h^{-1}$, respectively. Both active video games produced EE that was significantly greater than sitting watching television and seated video gaming. Indeed the energy expended from Dance Dance Revolution, Ultramix 2, was markedly increased in comparison to all the other activities including the other active video game.

The researchers concluded that active video games could have the capacity to increase children's EE especially as they appeared to retain the fun element provided by seated video games. The study however was not without several limitations as no explanation was provided for the selection of the active video games and there was no justification for the 15 min measurement period. Moreover, the order of the separate activities was not randomised thus reducing the rigour of the findings. Lanningham-Foster and colleagues (2006) also recognised that the laboratory environment was not reflective of children's true-to-life active video gaming and recommended that future research should be conducted in naturalistic settings (Lanningham-Foster et al., 2006).

Subsequent active video gaming studies corroborated the findings of Lanningham-Foster et al., (2006) that active video gaming elicited greater EE and so could be utilised to increase PA levels in children, but researchers persisted with laboratory protocols. Maddison and colleagues (2007) found the EE of 10-14 y children increased from 6.70±0.84 kJ·min^{-1} during seated video game play, to 12.14±1.26 kJ·min^{-1} when playing Sony© EyeToy, Groove and rose further to 27.21±7.12 kJ·min^{-1} with Sony© EyeToy, Knockout. The energy expended over a 5 min period from the latter game corresponded to a 400% increase above REE (Maddison et al., 2007). Similarly, Graf and colleagues (2009) with an extended play period of 30 min demonstrated that the combined EE of Dance Dance Revolution 1 with Dance Dance Revolution 2 and of Nintendo® Wii Sports bowling with Nintendo® Wii Sports boxing, was three times greater versus resting and walking on a treadmill at 2.6 km·h^{-1} and 4.2 km·h^{-1}. Walking on a treadmill at 5.7 km/h however was also found to produce three times the EE (Graf, Pratt, Hester, & Short, 2009). Yet with 26 boys aged 11.4±0.8 y, an 8 min bout of Nintendo® Wii Sports boxing produced EE equivalent to only low intensity PA (0.41±0.10 kJ· kg^{-1}· min^{-1} = 3 METS) when compared to resting, watching television, seated video gaming and playing other active video games (Nintendo® Wii Sports bowling and tennis and Nintendo® Wii Fit Skiing and Step). Indicating potentially that active video gaming should not replace outdoor play and sports (White, Schofield & Kilding, 2011).

Some studies included additional measures to establish whether active video games had additional health benefits. In a sample of 12, 9-12 y old children, it was established that the energy expended during 5 min bouts of active video game play with Sony Eye Toy Cascade, was 224% higher than seated video gaming and watching television. The greater energy elicited from this game was equivalent to moderate PA levels and the children's heart rates (HR) also increased by 59% (Straker & Abbott, 2007). In 18, 6-12 y old children, the active video games XaviX bowling and XaviX J-Mat (SSD Company Ltd, Shiga, Japan), were also shown to produce significantly greater EE and HR from 5 min game play. XaviX J-Mat elicited EE of 21.90±6.82 $J \cdot min^{-1}$ and a higher HR than XaviX bowling (Mellecker & Mcmanus, 2008). Nintendo® Wii Sports boxing, also elicited significantly greater EE (11.7±3.4 $ml \cdot kg^{-1} min^{-1}$) and higher HR responses (121.4±20.3 bpm), when compared with resting, seated video gaming and walking on a treadmill at 1.5 $miles \cdot h^{-1}$ in (n=24) 8-12 y old children (Penko & Barkley, 2010). When utilising the more recently developed Microsoft 360 Kinect console, 15, 9-10 y-old children played 15 min bouts of the low intensity active video game, Kinect sports Ten pin bowling and the higher intensity Kinect Sports 200 m hurdles and these were found to expend higher levels of EE established as 73.7±44.0 $J \cdot min^{-1} \cdot kg^{-1}$ and 294.6±77.2 $J \cdot min^{-1} \cdot kg^{-1}$, respectively. The arterial adaptations measured, demonstrated the games had beneficial effects on vascular function in the children however, the study sample size was very small (Mills et al., 2013).

Despite these positive findings, none of the previous studies provided justification for the timing of the bouts which ranged from 5, 8 and 15 min (Lanningham-Foster et al., 2006; Maddison et al., 2007; Mellecker & McManus, 2008; Straker & Abbott, 2007; White et al., 2011), to 30 min (Graf et al., 2009), or for the active video games utilised. There was no standardised protocol amongst researchers for active video gaming interventions or for the choice of games. Researchers tended to use the games that came free with the console and which required different movement patterns. Furthermore all of the above cited studies took place in the laboratory and measured EE by indirect calorimetry. Although considered a robust method this meant that it was necessary for the children to carry a backpack containing the calorimeter and wear a neoprene mask over their mouth and nose. The unfamiliar setting of the laboratory and somewhat inhibitive method of measurement for EE might have been stressful for the children and so could have reduced their EE. As such, the environment and protocols previously utilised were unlikely to resemble the real-life active video gaming practices of the individual populations.

2.6.4 Children's active video gaming physical activity

Some of the previously cited studies converted EE to PA (METS) to illustrate the effects of active video games on children's activity. Mitre and colleagues (2014) compared the movement made by 8-12 y old children whilst they played the same game (Sega Superstar Tennis) on Nintendo® Wii versus (vs) three seated video gaming consoles (Playstation 2-Sony Computer Entertainment, San Mateo, California, Xbox 360-Microsoft©, Redmond, WA, Nintendo® DS Lite-Nintendo Company Ltd., Kyoto, Japan) and television viewing with various on/off sound modes for 10 min each. Their movement was measured using a combination of inclinometers and accelerometers (Physical Activity Measurement System, PAMS). The children moved 50% more when playing the active video game on the Nintendo® Wii console which was subsequently calculated to be 3.05 METS and so just qualified as moderate PA levels (Mitre, Foster, Lanningham-Foster, & Levine, 2011). With Sony© EyeToy Groove, the PA levels of 10-14 y children during 5 min bouts were measured as being light intensity (2.3±0.3 METS) and with Sony© EyeToy, Knockout were established as moderate (5.0±0.8 METS) (Maddison et al., 2007). Similar PA levels were established by Graf and colleagues (2009) in male participants with regards to Dance Dance Revolution and Nintendo® Wii Sports bowling and boxing in a group of 10-13 y old children. The energy expended by the male participants from playing each active video game for 30 min, was shown to be equivalent to moderate intensity PA and for the majority of games was greater than that of girls (Graf et al., 2009). The higher PA intensity achieved in the aforementioned study by the boys during active video gaming, might reflect a greater level of engagement in the games. Indeed, the intensity of PA achieved by the 9-12 y old children whilst playing an internet based ten-pin bowling video game and XaviX bowling active video game for 1 h, followed by a choice of either a 1 h bout of seated internet-based running video game or the more vigorous XaviX J-Mat, was measured to be much higher in the boys. A greater proportion of boys attained vigorous levels of PA especially when playing XaviX J-Mat. Qualitative data also revealed the boys had more interest in active video games than the girls did, who were more likely to become frustrated and bored with active video gaming (Lam, Sit, & McManus, 2011).

The findings from all of the cited studies which examined either EE and (or) PA levels demonstrate that the level of energy expended or intensity of PA appeared to be dependent on gender, the active video gaming console being used, as well as the game being played. Recent research has indeed revealed that PA and EE are dependent on the gaming console (Simons, Vries, Jongert, & Verheijden, 2014). The EE and PA METS of six active video games (EyeToy, beach volleyball, ApartGame, Hitando, Lasersquash, Nintendo® Wii, Sports Tennis, Dance Dance Revolution Xerbike) were compared when played by 7-13 y old children. The researchers measured EE by indirect calorimetry and converted the values to PA METS. Nintendo® Wii Sports tennis, elicited light PA (2.7±0.6 METS) and vigorous levels were produced from Xerbike (6.1±1.4 METS). Xerbike however, is an active video game designed for use in public settings and not the home.

The findings of the cited studies, suggest that active video game play could be used to help increase PA levels or reduce sedentary time in children. Nonetheless, there must be a degree of caution as the majority of studies were conducted in the artificial confines of a laboratory and were not reflective of real-life paediatric active video gaming practices established thus far (O'Loughlin et al., 2012; Simons, Bernaards, et al., 2012). The short timings of the individual gaming bouts which ranged from 5, 10, 15 min (Maddison et al., 2007; Mitre et al., 2011), to 30 min (Graf et al., 2009), could also have meant the children were able to sustain a consistent level of PA and so moderate levels were more achievable. Furthermore, various active video gaming consoles and active video games were used and these too can have an effect on EE.

2.6.5 Measurement of active video gaming physical activity by accelerometry

To date, only a few of the studies discussed above have utilised accelerometers to assess the acute PA levels (METS) (Lam et al., 2011) or movement (Lanningham-Foster et al., 2006; Mitre et al., 2011) produced by children from active video game play. Yet the research by Lam and colleagues (2011) was the only study to solely employ accelerometers to measure the acute PA levels achieved by children, during active video gaming. The remainder used portable indirect calorimetry as well as accelerometers and inclinometers (Lanningham-Foster et al., 2006; Mitre et al., 2011). Indirect calorimetry as mentioned in section 2.6.3 requires the participant to wear a neoprene mask and carry a backpack containing the calorimeter, which could hinder the movement and thus affect the EE of the participant (Lamonte, Ainsworth, & Tudor-Locke, 2003), particularly that of a child.

In contrast, accelerometers are small, lightweight and unobtrusive devices that are unlikely to hinder movement. The more technologically advanced triaxial accelerometers are able measure movement in the vertical, anteroposterior (AP) and mediolateral (ML) planes. They also have the sensitivity to detect movement made when sitting and standing (Westerterp, 2009) and during frequent short bursts of vigorous intensity activity made in combination with moderate and low levels of PA (Rowlands, 2007). Accelerometers not only provide a measure of PA intensity but they also supply a record of the time spent at the different levels of intensity i.e. the min spent in sedentary, light, moderate, MVPA, vigorous and very vigorous activity . Furthermore, triaxial accelerometers can record accurately using various epochs (time-periods), which is especially useful when the activity being measured is light to vigorous (Nilsson, Ekelund, Yngve, & Sjostrom, 2002; Rowlands, 2007). The device is invariably placed on the right hip with an elasticated waist belt. Studies with children have established that hip placement is favoured over the lower leg and so it is considered the most comfortable location (Puyau, Adolph, Vohra, & Butte, 2002). Moreover, the majority of accelerometer research has favoured placement on the right hip and there is evidence to support this location as being the optimum site (Reilly et al., 2008; Ward, Evenson, Vaughn, Rodgers, & Troiano, 2005). Furthermore, when 11-17 y children played Nintendo Wii™ Sports tennis, right hip accelerometer placement was found to have a closer relationship with EE than when positioned on the upper body sites of the right or left wrist (Graves, Ridgers & Stratton, 2008).

Considering the technological capabilities of triaxial accelerometers, which enable the objective measurement of PA and the low participant burden in comparison to portable indirect calorimetry, they might provide a more realistic assessment of children's active video gaming PA. Furthermore, accelerometry will not only allow comparison with current public health recommendations for children's PA that are defined in this way (Reilly et al., 2008) but it will also enable an estimation of EE. It must be noted however that estimations from accelerometer data can result in an underestimation or overestimation of EE (Nilsson et al., 2008). However, considering the numerous laboratory studies, it would be pertinent to utilise accelerometry to enable the measurement of true-to-life active video gaming PA.

The study of active video gaming in children and adolescents to date has shown that acute EE and PA are increased by these games when compared with resting, watching television, playing seated computer games and walking. The increase in EE and PA nonetheless appears to be dependent upon the active video gaming console and game played. In the majority of the studies, portable indirect calorimetry was used to measure active video gaming EE and the weight of the equipment might have hindered the movement made by the children and subsequently disrupted activity behaviour. Furthermore the studies were predominantly conducted in the laboratory, an unfamiliar environment for children and one that does not reflect real-life. Paediatric short term active video gaming intervention research has reached a stage wherein assessment of EE and PA could be conducted outside of the laboratory with equipment such as accelerometers which enable studies to be more true-to-life. Furthermore, researchers have generally not provided justification for the gaming console, active video games or timing of bouts which might have enabled the children to sustain higher levels of EE and PA throughout. Future research should therefore establish the real-life active video gaming practices of the study population prior to investigation of the EE and (or) PA that can be elicited by active video games.

2.6.6 Assessment of energy intake and appetite during active video gaming

Amongst the abundant number of paediatric active video gaming laboratory studies which assessed the level of PA (METS) and/or amount of energy expended during game play, there has been only one study in children that included the assessment of EI. This is surprising considering the evidence that links sedentary screen based media with intakes of unhealthy foods and drinks in children. During sedentary screen based media usage it was apparent that there were negative associations with healthy food consumption and positive associations with unhealthy food intakes in children. Such EI behaviours could lead to positive energy status (Berentzen et al., 2014; Borghese et al., 2014; Chaput et al., 2011; Pearson & Biddle, 2011). It is feasible to hypothesise therefore that the undesirable EI behaviours taking place during sedentary based media use, could also be occurring during active video game play.

Mellecker and colleagues (2010) implemented a study of EI during enhanced active video game play. In two 1 h trials conducted on two separate days, 27 children aged 9-12 y played video games on XBOX® 360 when seated and 1 h of activity enhanced game play using the same console and games, whilst walking on a treadmill. During both gaming trials, the children were offered a snack of their choice *ad- libitum* and the amounts consumed at the end of each gaming scenario were measured. The addition of the motor component (treadmill) to the video game did not significantly affect the food intake of the children with 1.57 MJ·h and 1.60 MJ·h being consumed during seated and active video gaming, respectively. The findings thus suggested that the detrimental food intake behaviours established in sedentary screen based might also be a risk factor during active video game play. However, Mellecker and colleagues (2010) utilised a seated video game attached to a treadmill on which the children walked and not a genuine active console or game.

The first gaming study to include the measurement of appetite in combination with EI and gaming, was conducted with adolescents (15-17 y) using a seated video game and was described earlier in detail in section 2.5.7 (Chaput et al., 2011). The findings of this study demonstrated that acute EI following a 1 h bout of seated video gaming versus, 1 h resting produced greater EI without a significant alteration in subjective sensations of hunger, prospective food consumption and fullness (Chaput et al., 2011).

More recently, acute EI and subjective appetite sensations were measured during active video gaming, in 12-15 y old obese adolescent males (Chaput et al., 2015). Each participant took part in four 1 h trials which included a resting control, seated video game play (boxing, XBOX 360®), active video game play (boxing, XBOX 360®, Kinect) and a cycling exercise which was calibrated to elicit similar levels of moderate intensity activity to the active video game play (~65% of estimated $\dot{V}O_2$ max). Both the a*d-libitum* EI from a post gaming test lunch and subjective appetite sensations measured throughout the trials, were not found to be significantly different between the four different conditions.

In a longer-term study, Gribbon and colleagues (2015) compared the effects of 1 h of active video gaming (XBOX® 360, Kinect), with 1 h of resting, and 1 h of seated video game play (XBOX® 360) on acute EI and EE over 24 h and then 3 d in (n=26) 13-17 y old males. A randomised crossover within-subjects design was employed so that each participant completed each of the three above trials. Throughout each of the conditions, subjective appetite sensations which included hunger, prospective food consumption and fullness were measured using VAS. Although EE was significantly greater during the 1 h bout of active video gaming (1050 kJ) there were once again no differences in EI monitored over 24 h. The lack of difference in EI could also not be explained by the subjective appetite findings as these did not display significant alterations in sensations of hunger, prospective food consumption or fullness either. Furthermore, physical activity energy expenditure (PAEE) was monitored over the 24 h period and compensation for the extra EE was measured to have occurred and so energy balance was found to be similar between conditions. Immediately after the 24 h period therefore, the adolescents were therefore in a similar positive energy balance state (Gribbon et al., 2015). However, energy balance was restored after 3 d when EE compensated for the EI following the resting and seated video gaming trials. Yet Gribbon and colleagues (2015) utilised different methods to assess the EE of the participants. During the trials, indirect calorimetry was used to quantify EE in the laboratory but for the remaining 3 d of the intervention, accelerometry was the primary method of measurement (Gribbon et al., 2015). As such, the measurement of this variable was inconsistent, which might have caused discrepancy in the subsequent calculations of energy balance.

Both of the active video gaming studies and the research by Mellecker and colleagues (2010) did not establish the console and games most commonly played by their study populations, or indeed whether there were associated food intake behaviours, prior to intervention. Furthermore the studies of Chaput et al., (2015) and Gribbon et al., (2015) utilised indirect calorimetry as the method of measurement for EE which requires the participant to wear a face mask and carry a back pack which could inhibit movement. There is a need therefore for active video gaming appetite studies to be conducted in an environment which is more true-to-life and to utilise less inhibitive methods of EE. Furthermore considering there is evidence of EI during seated video gaming in children, food intake during active video gaming should also be explored.

2.6.7 Active video gaming energy intake and appetite assessment

When considering the positive findings to date that suggest active video gaming could be utilised to increase EE and PA levels in children, it is imperative that future research determines the real-life active video gaming practices of children. It should also establish whether food and (or) drinks are consumed during active video game play as this may counteract the energy expended and so result in a state of positive energy balance. Where feasible and depending on the measures taken, studies should also move out of the laboratory into an environment that is more representative of children's active video gaming. The measurement of both subjective (VAS) and objective appetite should be utilised to understand more clearly the hedonic and physiological mechanisms of the children's acute EI and appetite during active video gaming. To date, the only gaming study to include the measurement of an appetite hormone related to hunger, found that acylated ghrelin concentrations did not alter significantly (Chaput et al., 2011). No paediatric active video gaming studies have included the measurement of satiety-related hormones during active gaming. Future active video gaming research should therefore consider the measurement of these peptides.

CHAPTER III

THE REAL-LIFE ACTIVE VIDEO GAMING PRACTICES OF 7 TO 11 YEAR OLD CHILDREN

3.1 Introduction

In England, one third of children are either overweight or obese (Boodhna, 2013) and only 21% of boys and 16% of girls aged between 5 and 15 y are meeting the recommended 60 min of moderate PA per day (Niblett, 2015). A large proportion of their time also appears to be spent in sedentary activity, which is an average of 3.3 h on weekdays (excluding time spent at school), increasing to 4.2 h for boys and 4.0 h for girls on weekends (Niblett, 2015). In the UK, seated computer games are very popular with 100% of 6-10 y-olds regularly playing them (Pratchett, 2005) and so these might be partly attributable to the low levels of PA (Vandewater et al., 2004). Unhealthy food consumption and lower intakes of fruit and vegetables have been linked to seated media activities and so alongside low PA levels, these eating behaviours could contribute to positive energy balance in childhood (Pearson & Biddle, 2011; Strasburger, Jordan, & Donnerstein, 2010; D Thivel, Tremblay, & Chaput, 2012).

Recent laboratory-based studies have established that children are able to expend more energy and achieve higher PA levels [metabolic equivalents (METS)] through active video game play. It is possible for children to expend two to three times more energy by playing the Nintendo Wii™ Sports boxing game, in comparison to watching television or playing seated video games (Graf et al., 2009; Lanningham-Foster et al., 2009; Lanningham-Foster et al., 2006; Straker & Abbott, 2007). Children have also been able to attain PA levels of 5 METS which is equivalent to moderate levels of PA (Maddison et al., 2007; White et al., 2011). There is variability however in the levels of energy expended and PA attained and the console and game played are one possible reason for this. In terms of EE, Nintendo Wii™ Sports bowling has been shown to elicit 12.6 $kJ^{\cdot}\ h^{-1\cdot}$ kg (Graf et al., 2009) whilst an equivalent active video game (Ten Pin Bowling) played on Xbox 360 Kinect Sports, generated only 4.4 kJ· h^{-1}· kg (Mills et al., 2013). When playing Sony© Eye Toy Knockout (PlayStation® 2) boxing game, children have attained 5 METS (Maddison et al., 2007) yet with Nintendo Wii™ Sports boxing, PA was found to be an average of 3 METS (White et al., 2011).

As well as the various consoles and games, laboratory protocols have utilised gaming bouts that have ranged from 5 to 30 min duration (Graf et al., 2009; Mellecker & McManus, 2014; Mills et al., 2013). Such time-periods might not resemble children's real-life active video gaming activity particularly when considering the findings of two recent surveys of adolescent active video game play. In a Canadian active video gaming survey, game play was typically reported as being 50.5 min per bout (O'Loughlin et al., 2012). Whilst in The Netherlands, respondents revealed their game play was once per week for an average of 80 min (Simons, Bernaards, et al., 2012). It would be beneficial therefore for the average active video game play time of the population of interest, to be established prior to design of active video gaming intervention studies.

Active video game play might also be affected by socio-demographic characteristics. Indeed, there are data regarding the socio-demographic and active video gaming patterns for adolescents in The Netherlands and Canada (O'Loughlin et al., 2012; Simons et al., 2012). Being of a younger age was found to be associated with more frequent active video game play in Dutch adolescents (Simons et al., 2012) whilst in Canada, females were more likely to play these games than males (O'Loughlin et al., 2012). Information such as this in the UK on which to successfully inform intervention studies is lacking. There is also limited understanding of the reasons for children's active video game play, which might be dependent upon the study population. In New Zealand, children reported that the content of some active video games motivated their play (Dixon et al., 2010). They are also viewed as being a more social form of computer game than non-active video games as they enable game play against others. Indeed, Chinapaw and colleagues (2008) found an increased motivation to play when children played against other players which was believed to have reduced drop-out rate in an active video gaming intervention. It is also not known as yet, whether children play active video games alone or with others. Chinapaw and colleagues (2007) compared the effects of playing a dance simulation game alone at home, versus structured multi-player classes over a 12-week period, in 9-12 y-old children. The multi-player classes were found to have significantly lower participant drop-out from the study and the average active video game play time was found to be greater, than for those who played at home. These findings suggest that children have increased motivation to play active video games when they play with others (Chinapaw et al., 2008). Whether children play active video games with other people in their naturalistic setting should therefore be established.

As discussed in Chapter II, some recent paediatric studies have also established detrimental food intake behaviours during sedentary screen based media activities (Berentzen et al., 2014; Borghese et al., 2014; Marsh et al., 2013; Pearson & Biddle, 2011; Vandewater et al., 2004). Moreover when EI intakes of 9-13 y-old children were measured and compared during a laboratory-based bout of seated video gaming with an activity enhanced activity game, no differences were found in EI (Mellecker, Lanningham-Foster, Levine, & McManus, 2010). It would seem from the findings of the previously cited study, that EI during the activity enhanced active video game might counteract the energy expended. Unfortunately, active video gaming EE was not measured in the cited study, only REE. It would be pertinent therefore to explore whether children habitually consume foods or drinks in their naturalistic environment during active video game play and if so, what these foods and drinks are.

There is also limited understanding of the reasons for children's active video game play, and these might be dependent upon the population being studied. Thus far children have reported that the content of some active video games as a stimulus to play them (Dixon et al., 2010). They are also viewed as being a more sociable form of computer game since they enable game play with other individuals (Dixon et al., 2010; Simons, de Vet, et al., 2012). Information such as this in the UK is lacking and would prove beneficial for the design of ecologically valid interventions.

As discussed in Chapter II, effective approaches are needed to tackle children's low PA and unhealthy food intakes as these are the major contributors to positive energy balance in children (Cecchini et al., 2010; Kohl et al., 2012; Prentice & Jebb, 1995). Active video games might be one such approach however the majority of the research thus far has been laboratory-based and is not necessarily representative of young children's active video gaming practices in real-life. Therefore, the purpose of the present study was to establish the real-life active video gaming activity of 7-11 y children from Newcastle upon Tyne. To determine whether there are foods and (or) drinks consumed during active video game play and to explore why children play the games. Specifically, it aimed to examine, the access children have to active video gaming consoles and games, the favourite and most frequently played active video gaming consoles and active video games and the frequency and duration of play. To establish if children play active video games with other individuals and whether food and (or) drinks are consumed during game play and if so what these are. Lastly, to determine the reasons why, children play active video games.

3.2 Methods

3.2.1 Design

A cross-sectional survey study design was employed. The study was granted ethical approval by the University of Northumbria, Faculty of Health and Life Sciences Ethics Committee.

3.2.2 Questionnaire design

The Active Gaming Questionnaire was purposely developed to establish the real-life active video gaming activity and socio-demographics of the study population. Due to the young age of the children, the questionnaire was designed so that the parent answered the questions in consultation with their child. A copy of the Active Gaming Questionnaire is provided in Appendix A. Multiple choice questions were mainly utilised to reduce the burden of time upon participants and to increase response rate. Although a small number, were open-ended to enable the participants the freedom to provide unbiased and instinctive responses (Marshall, 2005; Oppenheim, 2003).

The questionnaire was informed by findings of active gaming studies that measured EE, PA or EI (Graf et al., 2009; Lanningham-Foster et al., 2006, 2009; Maddison et al., 2007; Mellecker & McManus, 2008). Section one of the questionnaire begins with some general questions relating to the type of console and game played and the kind of access the children had to them (questions 4 to 6). Remaining questions were developed using primary research. Questions 7 to 9 were based on those used in the International Physical Activity Questionnaire- Long Last 7 Days Self-Administered Format (Booth, 2000) to establish the frequency of the child's active video game play during the last 7 days. Questions 10 and 11 were informed from findings that children had increased motivation to play active video games when they played with an opponent (Chinapaw et al., 2008). The lack of difference found in children's EI between seated and activity enhanced gaming (Mellecker et al., 2010) and the unhealthy food intakes during sedentary screen based media informed questions 12 to15 (Berentzen et al., 2014; Borghese et al., 2014; Pearson & Biddle, 2011). Question 16 was developed from previously reported parental concerns of prolonged sitting during their children's seated computer game play and potential reduction in outdoor activity (Dixon et al., 2010). The socio-demographic questions (section one, questions 1 to 3; section two, questions 1 to 9) were informed by the Health Survey for England (2008) (Aresu et al., 2009), The General Lifestyle Survey (2008) (Ali et al., 2008) and a report on the 'Use and Abuse of Alcohol in UK University Sport' (Partington et al., 2010).

In terms of real-life active video gaming practices, the questionnaire was specifically designed to establish (i) the types of consoles and games the children played most frequently (ii) the type of access the children had to active video games (iii) the frequency, duration and mode of active video game play (iv) any food and/or drink consumption during active video gaming (v) the socio-demographics of the study population and (vi) the reasons for active video game play.

3.2.3 Pilot testing

Prior to data collection, pilot testing was carried out to confirm participant comprehension of questions and ensure the information acquired from the questionnaire met the study aims (Marshall, 2006; Oppenheim, 2003). Six parents and their children, who were aged from 7-11 y and who were known to play active video games, were invited by email to participate in the pilot study. A total of six parents and eight children (three boys and five girls) with an average age of 9.9±2.6 y agreed to take part. A copy of the Active Gaming Questionnaire was provided to each parent who was asked to complete it together with their child. As recommended by Oppenheim (2003) when developing a questionnaire, the participants in the pilot study were invited to provide feedback. They were asked to provide this by answering questions relating to the clarity of questions, the information and directions given, the design and layout, child comprehension, the suitability of questions and the timing of completion (Oppenheim, 2003).

As a consequence of the pilot study it was necessary to make a few amendments to the 'Active Gaming Questionnaire'. The revisions made included providing examples of active video games within the instructions on the cover page to provide greater clarity for respondents. The design and layout was improved and lastly, the wording of two questions (questions 10b and 15b) was changed for greater clarity. It was established that the timing required for completion was 10 min and this information was then included on the Parent/Main Carer Information Sheet.

3.2.4 Participants and data collection

The study population was acquired through primary schools located in Newcastle-upon-Tyne, a City within the North East of England (UK). To gain access to 7-11 y-old children, 15 primary schools were approached from areas within the city, the socio-demographics of which varied according to the Indices of Multiple Deprivation (a measure of relative deprivation levels) (Newcastle City Council, 2014). Written informed consent was obtained from three of the schools. Recruitment took place during the spring and summer school terms (from 10th April 2012 until 6th July 2012) either, during class time or at school events such as summer fayres. This enabled access to 310 children aged from 7-11 y and their parent, from which a purposeful sample who had access to play active video games, were invited to take part. Children who did not have access to play active video games, were excluded. Prior to data collection, written informed consent was obtained from 44 children and their parent or guardian from the three consenting schools. Forty four questionnaires were distributed to consenting participants in these schools and 40 were completed in full and returned.

3.2.5 Data analysis

The Statistical Package for the Social Sciences (SPSS; version 19, SPSS Inc., Chicago, IL) was used to analyse the responses to the closed questions within the Active Gaming Questionnaire. The responses to these questions created either dichotomous or categorical variables apart from question 10b (section 1), "When your child does play, how much time do they usually spend playing active video games?" which required a numerical answer in hours and min and was a continuous variable.

Shapiro-Wilk tests revealed all data were not normally distributed requiring the numerical responses for question 10b (section 1) to be log transformed. Descriptive statistics (frequencies and means) were calculated to establish the socio-demographics and the real-life active video gaming practices which comprised the consoles and games played most, the type of access to active video games, the frequency, duration and mode of the children's game play and to also determine whether foods and drinks were consumed during active video game play. Chi-square was used to check for significant associations between the frequency of play and whether foods and (or) drinks were consumed during play. Chi-square was also used to check for significant associations between whether parents believed active video games were or were not an alternative form of exercise for their child and if foods and (or) drinks were consumed during play. Differences in game play time according to whether food and/or drinks were consumed were explored using an independent samples t-test. Significance was accepted at $p<0.05$.

The responses to the two open-ended questions which established reasons for active video game play (section 1, questions 13 and 15b) were copied verbatim by the researcher and read through several times in order to become familiar with their content. Thematic analysis allowed the qualitative data to be categorized and organised in a meaningful way (Braun & Clarke, 2006). The method enabled common themes to emerge that were generic to the majority of participants and also some that were specific to a minority but nonetheless considered important (Patton, 2002). Such themes were regarded as essential to establish and understand participant responses and recognise the different experiences of active video gaming.

3.3 Results

3.3.1 Population characteristics

The majority of children who participated were boys (n=29, 72.5% *vs.* n=11, 37.5% girls). The mean $_{\pm SEM}$ age was 9.6±0.7 y. The children were mainly white (n=36, 90%), followed by Asian (n=3, 7.5%) and mixed ethnicity (n=1, 2.5%). Socio-demographic characteristics of the parent or main carer of each child participant are provided in Table 3.1.

3.3.2 Real-life active video gaming practices

In total, 95% of the children had access to an active video gaming console 'at home'. Fifty per cent of them had access to an active video gaming console only 'at home' whilst 45% not only had access 'at home' but also 'at a friend's home', 'at a relative's home' or elsewhere. Five per cent of the children had access only 'at a friend's home'. The most popular device played was the Nintendo Wii™, although a large proportion of the children (55%) played more than one type of active video gaming console. Nintendo Wii™ Sports was the favourite and most frequently played game, followed by Just Dance® and Microsoft© Kinect Adventures.

During the preceding 7-days, the average frequency of active video game play was '1-to 2-days', with Saturday being the day on which the majority of children (55%) usually played. The mean time of the children's active video game play was 81±50 min. Most children (97%) played active video games with other people, generally their 'brother/s or sister/s'.

Table 3.1. Socio-demographics of parent or main carer.

National deprivation bandings (number of child participants)				
0-10% (most deprived)	**10-20%**	**20-30%**	**30-50%**	**50-100% (least deprived)**
20	0	0	20	0

Total number of children in family						
1	**2**	**3**	**4**	**5**	**6**	**7**
8	12	6	6	5	1	2

Marital status of parent/main carer			
Single	**Married/co-habiting**	**Divorced/separated**	**Widowed**
5	34	0	1

Main source of family income			
Mother	**Father**	**State benefits**	**Other**
6	24	2	6

Main provider employment status			
Full time	**Part time**	**Studying**	**Not employed**
28	5	2	5

Approximate annual family income						
≤ £14,999	**£15,000-£19,999**	**£20,000-£29,999**	**£30,000-£49,999**	**£50,000-£99,999**	**>£100,000**	**Missing responses**
3	5	6	13	5	0	8

3.3.3 Food and drink intake during active video gaming

More than half (52.5%), of the participants ingested food and (or) drinks whilst they played. The food and drinks reported as being consumed by the children are listed in Table 3.2. The most popular foods were fruit (13%) and crisps (potato chips) (13%) and the most popular drinks were fruit flavoured juice (45%) and milk (10%). On average the children who ate and (or) drank whilst active video gaming, played for 82±68 min which was a significantly greater game play time than for those who did not eat or drink during play (57±32 min) (p<0.000). According to Chi-square, there were no associations between the consoles played (χ^2 =2.373, df=3, p=0.499), the type of access to active video games (χ^2 = 3.598, df=2, p=0.165), who they played the games with (i.e. mode) (χ^2 =0.498, df=3, p=0.919), the frequency of play (χ^2 = 4.293, df = 4, p= 0.368) and whether they ate and (or) drank during game play.

Table 3.2. Food and drink items consumed by participants (n=21) during active and seated video game play with foods offered *ad-libitum*.

Foods and drinks	**Proportion of children (%)**
Cordial	45.2
Crisps	12.9
Fruit	12.9
Milk	9.7
Biscuits	3.2
Flavoured water	3.2
Pizza	3.2
Salad	3.2
Sandwich	3.2
Water	3.2

3.3.4 Reasons for active video game play

When parents were asked if they believed active video games were an alternative form of exercise for their child, 65% responded “yes”, 32.5% “no” and 2.5% “don’t know”. Chi-square analysis revealed a significant association between whether parents considered active video games to be an alternative form of exercise and the consumption of foods and/or drinks during play (χ^2 = 12.494, df = 2, p=0.002). Those who reported that they did consider active video gaming to be an alternative form of exercise (65%), 73.1% of them responded that their children consumed foods and/or drinks during play. Whilst out of those who reported 'no', they did not consider active video gaming to be an alternative form of exercise (32.5%), 84.6% responded that their child did not eat and/or drink during play.

When asked to give a reason for their response ('yes'or 'no') as to why they believed active video gaming was, or was not an alternative form of exercise for their child, three themes emerged from the thematic analysis. These were '*physical effects*', '*outdoor games and play*' and '*environment*'.

3.3.4.1 Physical effects

'*Physical effects*' was the most common theme. Those who considered active video games to be an alternative form of exercise recognised beneficial effects when their child played them:

"They are good for hand-eye coordination which has made him a lot better at school but I'd rather see him play outdoors" [participant 5 (P5)].

"Because he's working up a sweat and is totally breathless" (P27).

In contrast, those who considered active video games were not an alternative form of exercise viewed the '*physical effects*' observed from active video game play as not so beneficial and believed they were insufficiently physically demanding:

"They're not because he's not getting any fresh air, it's not a team sport, there's no social interaction. [They're] not really physical and demanding and no real challenge" (P30).

"It's not proper exercise and [I] prefer him to play actual games" (P33).

3.3.4.2 Outdoor games and play

Active video games however, were generally not regarded as being a complete replacement for exercise and play outdoors with several parents reporting they preferred their child to play outside:

"[I] think the sports games are [an alternative form of exercise] but I still prefer my child to play outside" (P4).

"I consider exercise something done out of doors, not in front of a screen" (P12).

"[You] can't beat kicking a football around and doing other sports and being outside e.g. cycling and on a scooter" (P38).

3.3.4.3 Environment

Responses relating to the '*environment*' revealed that active video gaming was regarded as a good alternative when the weather restricted outdoor play. For some parents, they considered active video gaming as an alternative form of exercise due to a lack of safe green space in which to play:

"If for whatever reason e.g. weather, she can't get outside to play then this is the best alternative for being active" (P22).

"There's no park nearby and the neighbours complain about them playing outside" (P40).

In response to 'please give a main reason why your child plays active video games', four themes emerged from the thematic analysis and were labelled '*entertainment*', '*health benefits'*, '*unsafe environment'* and '*popularity':*

3.3.4.4 Entertainment

Entertainment was the most common theme to emerge. The majority of responses from parents and children about playing active video games as a recreational activity were positive:

"he likes running around, jumping about and making a noise" (P5).

"they're really fun, I really like jumping about" (P29).

"they're fun and challenging and I like to win against other people" (P21).

Quotes from some however introduced a somewhat negative perspective in relation to their use as a recreational activity:

"he has too much time and [he plays them] for something to do as he's too old for toys" (P2).

"he plays when he's bored" (P4).

3.3.4.5 Health benefits

Participants also conveyed how the movement facilitated by active video gaming and the types of games that they are able to play, combined entertainment with beneficial PA:

"she loves to dance and be active" (P22).

"[Child] (active video gaming) keeps me fit. [Parent] He likes indoor activity" (P27).

"likes the coordination and challenge" (P10).

3.3.4.6 Unsafe environment

Although in the minority, some participants raised the issue of neighbourhood safety which inhibited outdoor play as a reason for active video game play;

"the area is unsafe outside" (P3).

"she enjoys them and we live quite near a main road and not many friends live nearby" (P33).

3.3.4.7 Popularity

For some, the '*popularity*' of active video games amongst peers was the main reason their child played active video games:

"[It's' the] popularity of the games console and the games" (P36).

"because everybody else plays them and she prefers active video games to seated video games and enjoys them" (P33).

The popularity of active video games was also perceived as being negative which was initiated by their friends' game play:

"peer pressure, his friends play them so he does" (P38).

3.4 Discussion

The present study was the first to investigate the real-life active video gaming practices of UK children aged 7-11 y. The majority of responses received from the Active Gaming Questionnaire were from boys (boys 72.5% and girls 37.5%). The main findings revealed the habitual active video gaming activity of children within Newcastle upon Tyne (UK). The majority of children had easy access to active video gaming consoles and games, with the most popular being Nintendo Wii™ and the Nintendo Wii™ Sports game compendium. They tended to played them on 1 or 2 days per week, usually on a Saturday for an average of 69 min. During active video game play, the majority of children revealed they consumed food and (or) drinks. Those who ate and drank whilst active video gaming, typically played for longer (82±68 min), compared with those who did not eat and drink (57±32 min) during game play. Most parents regarded the games as an alternative form of exercise for their child giving reasons that related to the '*physical effects'* of them, '*outdoor games and play'* and the '*environment*'. Reasons for playing active video games included for '*entertainment*', '*health benefits'*, an '*unsafe environment'* and '*popularity'* of the games.

Similar to adolescent populations, the present study population was found to have easy access to active video gaming consoles and games, mostly at home (Dixon et al., 2010; O'Loughlin et al., 2012; Simons, Bernaards, et al., 2012). Like Canadian adolescents, the children preferred the Nintendo Wii™ console and the Nintendo Wii™ Sports game compendium (O'Loughlin et al., 2012). In previous laboratory-based studies this particular console and game has increased children's EE by two to three times, when compared with resting and sedentary media activities (Graf et al., 2009; Graves et al., 2008; White et al., 2011). Such EE is equivalent to exercising at a moderate level of $\geq$3 to <6 METS, as achieved in structured activities such as walking, jogging and basketball (Ainsworth et al., 2000).

Not unlike Dutch adolescents, active video game play was not frequent amongst the children in the present study population (Simons, Bernaards, et al., 2012). In the preceding 7-days, most of the children had played on only "1 or 2 days". Their game play was mainly on a Saturday for an average of 69 min, a duration comparable to Dutch adolescents which was reported as being for 80 min per week (Simons, Bernaards, et al., 2012). When considering the current PA guidelines for the UK (Almond et al., 2011) and assuming these children played active video games at a moderate intensity, if they did not engage in any other PA, their active video game play would only help to meet the recommended levels on 1 day per week. In agreement with the previous studies, the frequency of active video game play is therefore, insufficient to meet PA recommendations (Baranowski et al., 2012; Simons, Bernaards, et al., 2012). However, it is an activity that will assist in the reduction of time children spend being sedentary (Almond et al., 2011). One reason for such low frequency of active video game play in the present population could be a preference for '*outdoor games and play'* as also favoured by 13-14 y-old New Zealand boys (Dixon et al., 2010).

Most of the children played active video games against others which inferred that they viewed them as being a social activity, in common with Dutch children, adolescents and parents (De Vet et al., 2014; Simons, Bernaards, et al., 2012). They also used them as a form of '*entertainment'*, viewed them as being *popular* amongst their peers and observed '*physical effects'* from game play which they perceived as having '*health benefits'*. When considering the low levels of PA in children in England, these positive responses given as reasons for active video game play and the good accessibility of the games provide support for active video games to be employed to help meet current PA guidelines, as long as game play is more frequent. A positive outcome of more frequent active video game play could be that it aids in the reduction of potential health inequalities for some children who play them, due to a lack of safe or unsuitable green space in their surrounding environment (Brockman, Jago, & Fox, 2011; Lake & Townshend, 2013; Mitchell & Popham, 2008).

In the present study, children who consumed foods and (or) drinks during active video gaming reported playing active video games for significantly longer (82±68 min), in comparison to those who did not (57±32 min). As previous laboratory studies have shown that active video gaming can increase EE and PA levels in comparison to resting and sedentary activities, it is possible that this additional energy expended might be counterbalanced by EI. In contrast, EI could be in excess of the energy expended, as previously found in adults (Lyons, Tate, Ward, & Wang, 2012). The cited study found EI to be lower during active video gaming versus both television watching and seated video gaming, and EE was significantly greater. More energy was still ingested however, than expended during active video gaming (Lyons et al., 2012). This suggests that even though the children in the present study who reported eating and (or) drinking during active video gaming played for longer, their EI could still exceed the energy they expend. The consumption of food and (or) drinks whilst active video gaming might also be comparable to intakes during seated video gaming, as shown with 9-13 y children whilst they played a gaming device attached to a treadmill (Mellecker et al., 2010). The increased EE and PA might not be so beneficial when eating and drinking occurs during active video gaming. Instead, EI during active video gaming, similar to sedentary screen based media could potentially contribute to childhood positive energy balance (Berentzen et al., 2014; Borghese et al., 2014; Pearson & Biddle, 2011; Strasburger et al., 2010). Especially for those children whose *environment* is perceived as unsuitable for outdoor play (Mitchell & Popham, 2008).

Accordingly, the present study has established the most popular foods (crisps and fruit) and drinks (cordial and milk) (Table 3.2) consumed during active video gaming. In view of the literature linking sedentary screen based media and the recent active video gaming research, these foods and drinks should be used in future paediatric research to compare acute appetite sensations of hunger, prospective food consumption and fullness and EI during active and seated video gaming. Furthermore, the greatest number of responses to the Active Gaming Questionnaire were from boys aged 9.0±1.3 y, therefore in view of this and the screen based literature, it appears that boys in mid-to-late childhood are likely to be more at risk from the detrimental effects of screen based media. Accordingly, the population deemed to benefit most from the present study are boys aged 8-11 y. Further work should therefore explore the effects of active video gaming on acute appetite sensations and EI in 8-11 y old boys and so attempt to establish the mechanism for eating and drinking during game play. Parents could be advised whether specific types of food and drinks can be consumed during active and seated video gaming or whether EI should be restricted.

There were a number of strengths to the current study. It was the first study within the UK to examine the real-life active video gaming practices of 7-11 y-old children to explore reasons for active video game play. To do this a questionnaire (Appendix A) was purposely developed which included both structured and open-ended questions. Findings such as duration of game play, the most popular type of consoles and games played, whether they played active video games with others and food and drink consumption emphasise the importance for future research to determine these active video gaming practices prior to design of interventions. Moreover, they highlight the flaws in the study designs and subsequent PA and EE findings of the previous laboratory controlled studies.

In the present study the population was purposefully selected to be children who played active video games in a specific locality, namely the City of Newcastle upon Tyne. The City is located in the northeast of England and has a socio-demographic structure which is reflective of the UK population (White et al., 2004). The participants were recruited from schools situated within urban areas classified as having mid-to-high levels of social deprivation, due to study consent being refused by schools located in the lesser deprived parts of Newcastle upon Tyne (Newcastle City Council, 2014). Findings are bias therefore to children living in urban parts of Newcastle upon Tyne with mid-to-high levels of social deprivation and should not be generalised to other children living in rural locales, areas of low deprivation in the City, or other parts of the UK. The levels of deprivation in the present study population might have influenced participant choice in relation to the most played active video gaming console and game, the frequency and time of game play, whether they consume foods and (or) drinks during play and the type of food and (or) drinks they consume. However, when considering that the socio-demographic structure of Newcastle upon Tyne is typical of the UK (White et al., 2004), it could be anticipated that the present findings would be similar to other areas with mid-to-high levels of social deprivation. Although future studies would be needed to corroborate these findings. In addition, the ongoing advancements in gaming technology will likely lead to relatively frequent changes in the real-life active video gaming practices of 7-11 y children and other populations. For these reasons, the real-life active video gaming practices of other populations which undergo future investigation should be established first, prior to any intervention.

Despite the present study findings not being generalizable, many of the real-life active video gaming practices were found to have similarities with other paediatric populations (Dixon et al., 2010; O'Loughlin et al., 2012; Simons, Bernaards, et al., 2012). Furthermore, some of the survey questions could not be validated as they were specifically developed for the current study. The questions were however, pilot-tested in a matched population to check their understanding and that responses met with study aims. All participants provided self-completed responses to the two open-ended questions although some of these were noted by the researcher to be short and cryptic. The qualitative information obtained could have been enhanced had interviews been conducted, enabling interviewers to probe for more comprehensive information. However, more participants could be recruited by using a questionnaire approach and it was not a primary aim of the present study to examine in-depth responses from a small population. It was more important to gather specific information regarding the real-life active video gaming practices of children.

In conclusion, the children in this study, the majority of whom were boys, typically played Nintendo Wii™ Sports, once or twice per week for 81±50 min. This frequency of active video game play would need to increase per week to help meet the recommended levels of PA. The duration of game play was also increased by 25 min when food and (or) drinks were consumed whilst gaming, which might have implications on childhood energy balance. The information acquired from the present study provide a good basis on which to inform future active video gaming intervention study designs, which when logistically possible can be conducted in naturalistic environments.

When considering the findings of the present study, it seems a logical next step for further research to utilise the habitual active video gaming practices of young children established here. This information should be used to inform the study design of future active video gaming interventions in more naturalistic settings, albeit depending on the measures required and when logistically possible. In doing this, the most popular console and game could be utilised, which for the child population studied here was the Nintendo Wii™ and Nintendo Wii™ Sports. The PA levels (METS) of active video gaming could be measured objectively using accelerometry, as previously demonstrated by Baranowski and colleagues (2012). Moreover, given the high proportion of children found to consume foods and (or) drinks during active video game play, and the associated extended gaming time reported here, it would be appropriate to explore the quantities of the most popular foods and drinks ingested whilst playing and also utilise the longer game play time. The present study has established the real-life active gaming practices of 7-11 y children in a subsample of children from Newcastle upon Tyne. In doing this, it has filled a gap in the active gaming literature and informed the next phase of the present work.

CHAPTER IV

THE ACUTE SUBJECTIVE APPETITE AND ENERGY INTAKE OF 8 TO 11 YEAR-OLD BOYS DURING ACTIVE AND SEATED VIDEO GAMING

4.1 Introduction

There is evidence that the prevalence of seated media activities, including television viewing, computer use and computer game play increases, as PA decreases in children during mid-to-late childhood (Huang et al., 2013). The increased seated media activity appears to be particularly predominant in boys and seems to reduce the time they spend undertaking sports and other physical activities (Atkin et al., 2013; Hands et al., 2011). Links have also been made between high screen use and an increase in child overweight and obesity (Hands et al., 2011). In addition, there are positive associations between television viewing and computer use and the spontaneous intake of unhealthy foods and drinks, in children aged $\leq$11 y (Borghese et al., 2014; Pearson & Biddle, 2011; Vandewater et al., 2004). The combination of sedentary screen based media use, the EI during it and the lack of PA could therefore, be a major contributor to child positive energy balance.

Active video games require physical interaction and movement from the player and so might present an appealing way to increase children's PA and thus offset spontaneous EI (Mathieu & Kakinami, 2011). Recent laboratory-controlled studies have indeed established that active video game play can increase children's EE threefold, in comparison to sedentary pursuits (watching television or playing seated video games) (Graf et al., 2009; Lanningham-Foster et al., 2009; Lanningham-Foster et al., 2006; Straker & Abbott, 2007). Moreover, some active video games such as 'EyeToy Knockout' (PlayStation 2) (Sony, Tokyo, Japan) have been shown to elicit an intensity of 5 METS (moderate PA) (Maddison et al., 2007; White et al., 2011). Such findings suggest that active video games have the potential to contribute to the achievement of energy balance.

To the authors' knowledge, Mellecker and colleagues (2010) were the first to explore acute EI during an active video gaming intervention study with children. However, the active gaming device was a seated video game attached to a treadmill. During two, 1 h laboratory gaming sessions, snacks were made available *ad-libitum* to 9-13 y children. No significant differences in snack consumption were found between the seated and activity enhanced video gaming conditions and the mean energy EI was 66% above resting levels (Mellecker et al., 2010). This suggests that the additional PA elicited by the active video gaming bout, did not actually offset EI in this group. No measures of appetite were explored however and EE was not estimated, failing to provide insight into any potential mechanisms for these changes in EI.

More recently, both appetite and EI have been explored in seated and active video gaming studies with paediatric groups. In relation to seated video gaming, no differences were found in sensations of hunger, prospective food consumption or fullness, when compared with resting conditions in both male adolescents (aged 15–17 y) (Chaput et al., 2011) and boys (aged 9-14 y) (Branton et al., 2014). In the adolescent group, EE and *ad-libitum* EI were significantly higher than after resting (Chaput et al., 2011). Whilst the food intake of the younger boys (9-14 y) was lower after 30 min of seated video game play versus the identical period of resting (Branton et al., 2014). However, both of the former studies, provided food and drinks following the seated and resting bouts and not during them. Nonetheless, both groups were found to be in positive energy balance at cessation of the bouts.

Similar to the previously cited work (Branton et al., 2014; Chaput et al., 2011) , Marsh and colleagues (2013) compared the appetite sensations and EI of 9-13 y boys during seated video gaming and television viewing. In contrast to the former studies (Branton et al., 2014; Chaput et al., 2011), food and drinks were offered *ad-libitum* throughout the conditions. Although no differences were found in sensations of hunger, prospective food consumption or fullness, the EI of the boys was considerable during both seated video gaming (2.91±0.31 MJ) and television viewing (3.44±0.31 MJ) (Marsh et al., 2013), which resulted in positive relative energy balance.

At the time of writing, the effects of active video gaming on acute paediatric appetite sensations and EI have only been explored in 12-15 y (Chaput et al., 2015) and 13-17 y (Gribbon et al., 2015) males. In relation to sensations of hunger, prospective food consumption and fullness, no differences were found between active video gaming and the various comparison trials, which were sitting, cycling (Chaput et al., 2015), resting and seated video gaming (Gribbon et al., 2015). The cited studies also found no differences in self-reported sensations of hunger, prospective food consumption and fullness between active video gaming and any of the other named conditions. Furthermore, the EI of the adolescents did not significantly alter between trials although it should be noted that food was offered *ad-libitum* following the bouts and not during them (Chaput et al., 2015; Gribbon et al., 2015).

Appetite sensations and EI have not as yet been measured in combination during active and seated video gaming, in children. Furthermore, all of the previous studies have been strictly laboratory-controlled and so the gaming protocols employed did not resemble the real-life active video gaming practices of young children. In Chapter III, the real-life active video gaming practices of 7-11 y-olds from Newcastle upon Tyne (North East, England, UK) were established, to enable active video gaming interventions to be designed that are representative of children's habitual gaming practices. Therefore the primary aim of the present study was to explore the acute EI and subjective appetite sensations of hunger, prospective food consumption and fullness, during real-life active video gaming and seated video gaming, in 8-11 y boys. Secondary aims were to measure PA, estimate both EE and relative energy balance and establish time to eating onset.

4.2 Materials and methods

4.2.1 Study design

A randomised, cross-over design was used to compare acute EI and appetite sensations of 8-11 y-old boys during four gaming bouts each separated by 1 week, utilising methods identified in a previous study. The four gaming bouts were: 1) 90 min seated video gaming, no food or drink offered; 2) 90 min active video gaming, no food or drink offered; 3) 90 min seated video gaming with food and drink offered *ad-libitum*; 4) 90 min active video gaming with food and drink offered *ad-libitum*.

The study was approved by the University of Northumbria, Faculty of Health and Life Sciences Ethics Committee. Written informed consent was obtained from both the child and their parent (or main carer) prior to data collection.

4.2.2 Recruitment

To enable recruitment of 8-11 y boys and for logistical reasons, primary schools within Newcastle upon Tyne (North East England, UK) that were within a 6 mile radius of the university, were identified. Head Teachers of suitable primary schools were approached by letter. Each letter was followed-up with a phone call or email so that a meeting to explain the study in greater detail could be arranged with Head Teachers. Meetings were arranged with two Head Teachers who both consented to the recruitment of their male pupils aged 8-11 y and for the study to be implemented on the school premises, as an after-school club. During meetings with the 8-11 y boys, the study was explained and a demonstration of the gaming equipment and games was provided. The meetings enabled the researchers to distribute a total of 60 recruitment packs to all eligible boys in the two schools who expressed an interest in participating. They were asked to take the recruitment pack home to their parent (or main carer) to read. The pack contained a letter addressed to their parent/main carer with a full explanation of the study and consent forms for them and their child to sign and return to school. Boys were excluded from participating when injury or illness prevented them from being able to play active video games or if they had intolerances or allergies to the foods provided in the study. Signed consent was received from 22 boys, although one boy dropped out as his parents were unable to standardise his EI prior to the gaming bouts. A total of 21 boys took part in the study.

4.2.3 Anthropometric assessment and familiarisation

Prior to the first gaming bout, the researchers visited the school to meet the children and their parent or main carer. The purpose of the visit was to carry out anthropometric assessment. For each boy, the stature and seated height as measured to the nearest 0.01 m using a Harpenden Portable Stadiometer (Holtain Limited, Pembs, UK). Body mass was measured to the nearest 0.1 kg using portable SECA scales (SECA United Kingdom) whilst wearing light clothing. Waist circumference was measured to the nearest 0.01 m with a non-elastic flexible tape at each boy's natural waist whilst standing as a measure of central adiposity (McCarthy, Jarrett, & Crawley, 2001).

During this visit, the boys (and where applicable their parent or main carer) were familiarised with the gaming consoles, the games, the gaming session format, the self-reported weighed food diaries and visual analogue scales (VAS) used to measure appetite sensations. A demonstration of the right hip placement of accelerometers (Actigraph LLC © GT3X+) to enable measurement of PA during the gaming bouts was also provided. The boys were asked to complete a food preference questionnaire during the familiarisation session to ensure they did not dislike any of the foods and drinks offered during the study. The boys were also familiarised with the appetite VAS and completed the Dutch Eating Behaviour Questionnaire for children (DEBQ-C), as a measure of dietary restraint (van Strien & Oosterveld, 2008).

4.2.4 Study protocol

Each boy was provided with a self-report, weighed food diary and a set of food weighing scales (Salter©, Kent, UK) to use prior to all intervention days. With the help of their parent they were asked to weigh and record the amounts of all foods and drinks consumed from 17:00 the preceding evening, until after breakfast on the morning of each intervention day. A photocopy of this food record was provided to each parent and they were asked to replicate their child's food and drink intake prior to each gaming bout on three further intervention days. The boys were also asked to abstain from all PA from 17:00 the preceding evening and from physical education lessons at school.

The boys attended school as normal at 0855 h. If any of the boys usually consumed a snack during their morning break (1040 h), they were provided with this in each intervention week by the research team. The snack was dependent on the personal food intake of the boy and was the same each week. At lunchtime (1200 h) in the first week, each boy consumed a packed lunch prepared by the research team which comprised their preferred food and drink items. The food and drink items consumed were weighed and recorded by the research team so that an identical lunch could be provided in each of the three subsequent visits.

4.2.4.1 Gaming interventions

The design of the individual gaming bouts was based on published data which described the real-life active video gaming practices of 7-11 y children from Newcastle upon Tyne (Allsop, Rumbold, Debuse & Dodd-Reynolds, 2013). Thus the gaming console used for the active video gaming bouts was Nintendo Wii™ and the game used was Nintendo Wii™ Sports tennis. The seated video game utilised was 'Mario and Sonic at the London 2012 Olympic Games', which was played on the handheld Nintendo© 3DS device. Each of the four gaming bouts previously described in section 4.2.1, was separated by 7 days and took place on the same school day of each week over four consecutive weeks. The gaming bouts were implemented on the premises of the two consenting primary schools as an after-school club. The gaming bouts were timed over 90 min and began at 1530 h and ended at 1700 h, when the boys were collected by a parent (or main carer). The boys were tested in groups of four so that two of boys played Nintendo Wii™ Sports tennis, on the Nintendo Wii™ , whilst two played Mario and Sonic at the London 2012 Olympic Games' on the Nintendo 3DS. The pair who were assigned to the Nintendo Wii™ played together against the computer, whereas the two boys who played the Nintendo 3DS played individually against the computer. In doing this, peer influence related to winning or losing was avoided along with any subsequent effects on EI. As far as possible, the boys were paired according to year group. They were randomly assigned to a different gaming bout every week, so that by the end of 4 weeks they had completed each of the four trials.

On the intervention days, the boys were met in a designated room at school at 1515 h by two members of the research team. On arrival, they were able to relax until 1530 h when they completed the VAS for hunger, prospective food consumption and fullness. Following this, an accelerometer (Actigraph LLC© GT3X+) which had been programmed to record PA counts in 10 s epochs was then placed on the right hip of each boy. The foods and drinks were positioned at a station designated to each boy. The boys were asked to take food and drinks from their designated station only. The boys were then requested to commence gaming for 90 min during which they completed further appetite VAS at 30 min and 60 min, at the end of gaming (90 min) and also at 15 and 30 min post gaming.

4.2.5 Physical activity assessment and energy expenditure

During every gaming bout, the PA levels of each boy were measured by accelerometry using an Actigraph© LLC, GT3X+, worn on the right hip (Rowlands, 2007). The boys wore the accelerometer from when the gaming bouts commenced at 15:30 until they ended at 17:00 on each intervention day. At the end of every gaming session, the recorded data was downloaded utilising Actilife 6 data analysis software and interpreted using the recommended child-appropriate activity cut-off values of Evenson, Catellier, Gill, Ondrak and McMurray (2008). For conversion into metabolic equivalents (METS) the activity counts were integrated into 60 s epochs. The following MET thresholds were used to categorise the data as these are recommended for use with children to describe the intensity of the activity: sedentary < 1.5 METs; light 1.5 to < 4 METs; moderate 4 to < 6 METs; vigorous > 6 METS (Trost et al., 2010). In addition, the mean time (min) spent sedentary, in light PA and MVPA during each 90 min gaming bout, was calculated for each boy.

For each boy, Henry's body mass, stature and sex-specific equations were used to calculate basal metabolic rate (BMR) (Henry, 2005). Energy expenditure was then calculated as recommended by Ridley, Ainsworth and Olds (2008), as follows; METS x BMR (MJ·min·d) x 90 min gaming = MJ. This method of EE estimation was considered the most appropriate as it accounts for age (y), sex, body mass (kg) and stature (m), unlike other prediction equations which utilise only one or two of these physiological characteristics (Corder et al., 2007; Nilsson et al., 2008; Trost et al., 2010; Trost, Way, & Okely, 2006).

4.2.6 Energy intake and relative energy balance

The food and drink items provided during the gaming sessions were also based on the previous findings (Allsop et al., 2013) and are displayed with respective macronutrient values in Table 4.1. Apple was selected due to it being one of the most popular fruits consumed in England Institute of Food Research, 2014). All food items were pre-weighed by the researchers to the nearest gram using electronic portable scales (Salter©, Kent, UK) and all drinks were measured to the nearest millilitre. They were all numerically coded by the researchers and just before the gaming commenced, were placed at a station designated to each individual boy. The crisps and apple were placed in clear plastic bags and the milk and squash were placed in coloured drinks bottles so that volumes were not identifiable. The foods and drinks were offered *ad-libitum* and the researchers noted each bag or bottle taken by the boys and at the end of each gaming session, anything left over was weighed or measured so that amounts consumed could be calculated and recorded. If further food and drink items were required during the gaming bouts, additional portions were served as described in Table 4.1.

To estimate EI from the food and drink items served, nutrition information was obtained from individual food labels, an online resource (www.asda.com) and MicroDiet (Downlee Systems©, Derbyshire, UK). When the gaming bouts with food and drinks commenced, the time of the first eating episode for each boy was recorded. For each boy, EE was subtracted from the amount of energy consumed during each 90 min gaming bout in which foods and fluids were offered *ad-libitum* to calculate relative energy balance. Relative energy balance was calculated after each gaming bout as follows;

Gaming EI (MJ) – Gaming EE (MJ).

Relative energy was calculated to include EI of the test meal as follows;

Gaming EI (MJ) + test meal EI (MJ) – gaming EE (MJ).

Table 4.1. Serving size, total energy and macronutrient values of food and drink items served during the gaming sessions with food and drink.

Food or drink	Serving size	Energy (MJ)	Carbohydrate (g)	Fat (g)	Protein (g)
Apples ("Royal Gala" raw, sliced and cored)	130 g	0.26	15.60	0.13	0.52
Walker's© ready salted crisps	50 g	1.10	25.75	15.95	3.05
Semi-skimmed milk	250 mL	0.52	12.00	4.50	9.00
"Robinson's" apple and blackcurrant squash (no added sugar)	350 mL (1:8 dilution)	0.19	2.50	0.00	0.00

4.2.7 Subjective appetite

Sensations of hunger, prospective food consumption and fullness were assessed using paper-based VAS (Flint, Raben, Blundell, & Astrup, 2000). Visual analogue scales assess appetite states by asking participants "How hungry do you feel now?", ''How much would you like to eat now?'' and "How full do you feel now?" In response to these questions, participants are required to place a vertical mark along a horizontal line of 100 mm length. At the opposing ends of the horizontal line there are two extreme states of hunger, prospective food consumption and fullness. The hunger statements are, "very hungry" (at 0 mm) and "not at all hungry" (at 100 mm), for prospective food consumption they are, "a lot" (0 mm) and "nothing at all" (100 mm) and for fullness are "very full" (0 mm) and "not full at all" (100 mm). The boys were asked to complete the VAS immediately before they commenced gaming (0 min: 15:30), at 30 min (16:00) and 60 min (16:30) during the gaming session, at the end (90 min: 17:00) and at 15 min (17:15) and 30 min post gaming (17:30).

4.2.8 Statistical analysis

IBM© Statistical Package for Social Sciences© (SPSS) (version 22.0, SPSS Inc., Chicago, Illinois) was used for all statistical analyses. Means $_{\pm SEM}$ are presented for all data which was checked for normality using Shapiro-Wilk. When data was not normally distributed, log transformation was performed and outliers removed if necessary. One-way repeated measures analysis of variance (ANOVA) was used to detect differences between mean baseline values for sensations of hunger, prospective food consumption and fullness, mean PA and EE and time (min) spent sedentary, in light PA and MVPA for all gaming bouts. Further one-way repeated measures ANOVA were used to detect differences between the time (min) spent in sedentary activity, light PA and MVPA during all gaming bouts. A Bonferroni correction was made when significant differences were identified. Appetite VAS ratings for subjective hunger, prospective food consumption and fullness were calculated as time-averaged area under the curve AUC x 120 min for all gaming trials (15:30 – 17:30), to establish whether there was a specific effect of gaming. For the two gaming bouts in which food and drinks were offered *ad-libitum*, paired t-tests were used to establish differences in EI (MJ), relative energy balance (MJ), time to eating onset (min), macronutrient intake and the individual foods and fluids consumed. For significant differences found in the t-test analyses, Cohen's *d* effect size was calculated and interpreted against the effect size categories of ≤ 0.20 = small effect, ~ 0.50 = moderate effect, and ≥ 0.80 = large effect (Cohen, 1992). Statistical significance was set at $p<0.05$ for all analyses.

4.3 Results

4.3.1 Population characteristics

The mean $_{\pm SEM}$ age of the boys was 9.8±0.9 y, stature 1.39±0.01 m; body mass 35.5±1.7 kg; waist circumference 64.3±1.8 cm and BMI 18.4±0.8 kg/m^2. According to UK age and sex-specific BMI centiles (Cole et al., 1995) the majority of the boys were classified as having a healthy body mass (71.4%), 14.3% were classified as overweight and 14.3% as obese. The mean maturity offset was -1.1±0.2 y, indicating that the boys were an average of 13.2 months from peak height velocity. In addition, all boys were identified as being unrestrained eaters according to the Dutch Eating Behaviour Questionnaire (van Strien & Oosterveld, 2008) with a mean $_{\pm SEM}$ dietary restraint score of 1.7±0.10 categorized as being average for boys of this age 1.53±0.06.

4.3.2 Physical activity levels and energy expenditure

Active video gaming elicited only light PA and seated video gaming was sedentary (Table 4.2). The PA level (METS) during active video gaming with food were significantly greater than seated video gaming with food (p<0.001, moderate effect size 0.6). Likewise the PA (METS) during active video gaming without food were significantly greater than seated video gaming without food (p<0.001, moderate effect size 0.7). As expected, no differences were found between active video gaming with or without food (p=1.000) and between seated video gaming with or without food (p=0.389) (Table 4.2).

These results were confirmed when EE (MJ) was estimated from PA. Active video gaming EE with food was significantly greater than seated video gaming with food (p<0.001, moderate effect size 0.7). Similarly, active video gaming EE without food was significantly greater than seated video gaming without food (p<0.001, large effect size 0.8). As expected, no differences were found between active video gaming with or without food (p>0.05) and between seated video gaming with or without food (p>0.05) (Table 4.2).

Table 4.2. Mean $\pm_{SEM}$, PA (METS), energy expenditure (EE) (MJ), energy intake (EI) (MJ), relative energy balance (MJ), time to eating onset (min) for all boys (n=21) for each gaming bout.

	Seated no food		Active no food		Seated with food		Active with food	
	Mean	SEM	**Mean**	SEM	**Mean**	SEM	**Mean**	SEM
PA levels (METS)	1.38	0.07	2.14[a]	0.10	1.49	0.74	2.08[b]	0.90
EE (MJ)	0.43	0.02	0.69[c]	0.03	0.51	0.03	0.66[d]	0.03
EI (MJ)					2.88	0.26	2.30	0.28
Relative energy balance (MJ)					2.42	0.25	1.64[e]	0.28
Time of eating onset (min)					6.90	1.52	17.10[f]	4.00

[a] PA (METS) were greater during active video gaming without food than seated video gaming without food (p<0.001).
[b] PA (METS) were greater during active video gaming with food than seated video gaming with food (p<0.001).
[c] EE was significantly greater during active video gaming without food than seated video gaming without food (p<0.001).
[d] EE was significantly greater during active video gaming with food than seated video gaming with food (p<0.001).
[e] Relative energy balance was significantly lower when active video gaming with food than when seated video gaming with food (p=0.031).
[f] Time of eating onset (min) was significantly longer during active video gaming with food (p=0.017).

4.3.2.1 Time spent sedentary, in light and moderate to vigorous physical activity

When time spent sedentary, in light and MVPA was examined, it was revealed that the boys spent less time being sedentary during active video gaming without food (22.38±4.15 min) and active video gaming with food (25.48±3.75 min), than when both seated without food (64.29±3.99 min) and seated with food (55.29±4.35 min) (all $p<0.001$). More time was spent in light PA during active video gaming without food (66.05±3.95 min), than in the other three gaming bouts (active video gaming with food 63.33±3.60 min; seated video gaming without food 25.33±3.90 min; seated video gaming with food 34.67±4.35 min) (all $p<0.001$). More time was spent in light PA during active video gaming without food, than when active video gaming with food ($p<0.001$) and more time was spent in light PA when seated video gaming with food, than when seated gaming without food ($p<0.001$). For time spent in MVPA, active video gaming without food was significantly greater than seated video gaming with food ($p=0.019$) (Table 4.3).

Table 4.3. Mean $\pm_{SEM}$ time spent in sedentary, light PA and MVPA for all boys (n=21), for each gaming bout.

	Seated no food		**Active no food**		**Seated with food**		**Active with food**	
Time (min) spent	**Mean**	SEM	**Mean**	SEM	**Mean**	SEM	**Mean**	SEM
Sedentary	64.29	3.99	22.38[a]	4.15	55.29	4.35	25.48[a]	3.75
Light PA	25.33	3.90	66.05[b]	3.95	34.67	4.35	63.33	3.60
MVPA	0.38	0.18	1.57[c]	0.50	0.05	0.05	1.19	0.46

[a] Significantly, less time was spent being sedentary when active video gaming with and without food, than seated video gaming with and without food (all $p<0.001$).
[b] Active video gaming without food significantly increased the time spent in light PA, in comparison to active video gaming with food and seated video gaming with and without food (all $p<0.001$).
[c] Time spent in MVPA was significantly greater during active video gaming without food, than when seated gaming with food ($p=0.019$).

4.3.3 Energy intake, relative energy balance, time to eating onset, macronutrient intake and individual foods consumed.

When foods and drinks were offered during the 90 min bouts, a paired t-test revealed no significant differences between active and seated video gaming in total EI (MJ) ($p=0.238$). Despite the lack of differences detected in EI, the average time to eating onset (min) was significantly longer during active video gaming with food, in comparison to seated video gaming with food (*p*=0.017, small effect size 1.0) (Table 4.2). When EE (MJ) was subtracted from EI, there was a significant difference between the mean relative energy balance for active and seated video gaming (*p*=0.031, small effect size 0.3) (Table 4.2).

In terms of macronutrients consumed, no significant differences were detected in intakes of CHO (active 47.5±1.5 % versus seated 45.9±1.3 %, *p*=0.553), fat (active 40.7±1.7 % versus 42.6±1.2 %, *p*=0.649) or protein (active 11.8±1.1 % versus 13.5±1.4 % *p*=0.185,) as a percentage of total EI (MJ), between the 90 min active and seated video gaming bouts (Figure 4.1).

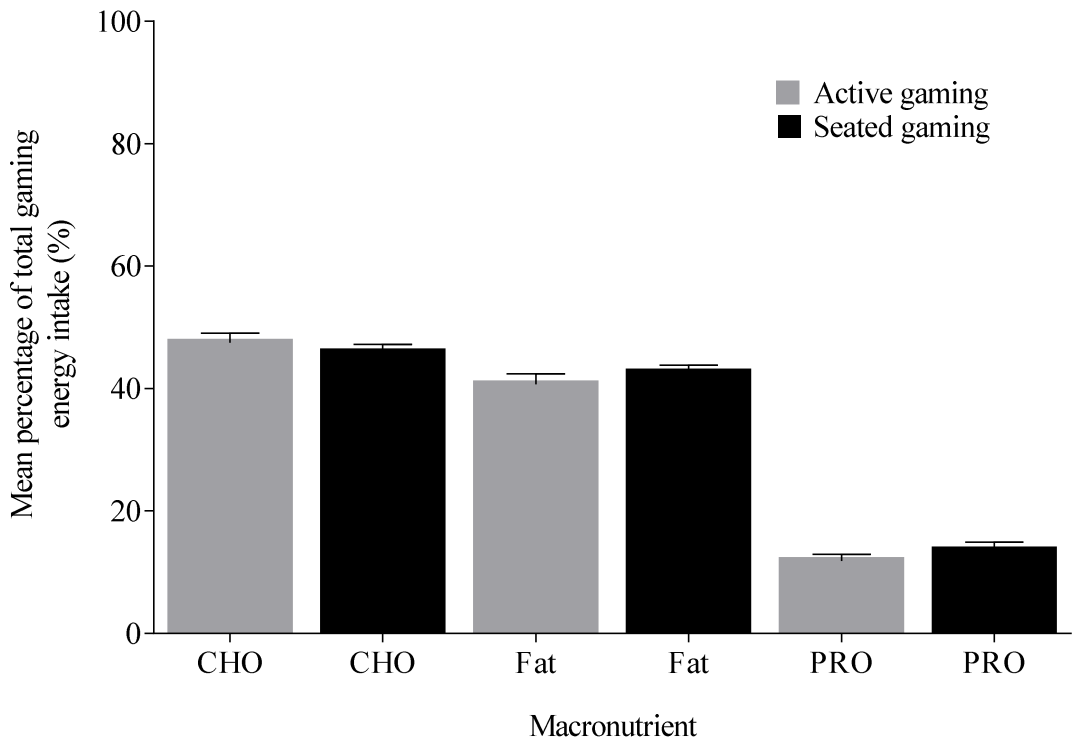

Figure 4.1. Mean amount of macronutrients ingested during gaming for all boys (n=21) as a proportion (%) of total EI. No significant differences in intakes of CHO (*p*=0.553), fat (*p*=0.649) or PRO (*p*=0.0.185) during active and seated video gaming.

When amounts of individual foods (crisps and apple) and fluids (fruit squash and semi-skimmed milk) consumed during the gaming bouts were examined, the majority of EI was due to crisps. A paired t-test revealed significantly fewer crisps were eaten during active versus seated video gaming (active 1.25±0.21 MJ versus seated 1.70±0.26, p=0.011, small effect size 0.4). No significant differences were detected in intakes of apple (active 0.26±0.05 MJ versus seated 0.27±0.05 MJ, p=0.579, fruit squash (active 0.10±0.02 MJ versus seated 0.13±0.03 MJ, p=0.967) or semi-skimmed milk (active 0.55±0.11 MJ versus seated 0.74±0.09 MJ, p=0.184) (Figure 4.2).

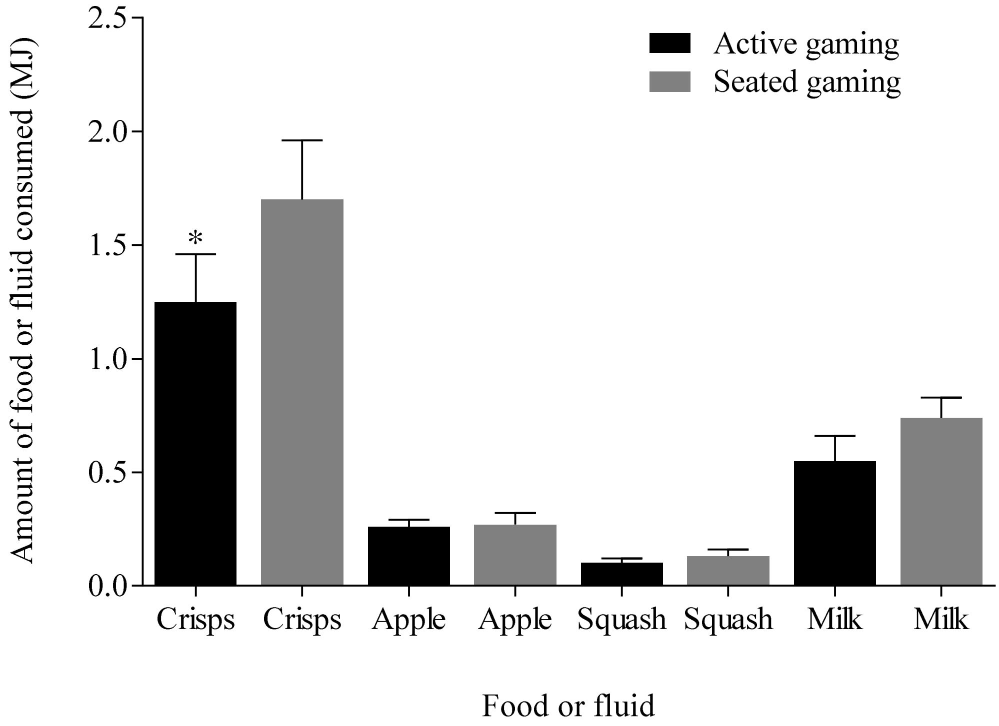

Figure 4.2. Mean amounts of foods and fluids consumed during active and seated video gaming for all boys (n=21). The amount of crisps consumed was significantly lower during active video gaming in comparison to seated video gaming (p=0.011). No significant differences were detected in intakes of apple (p=0.579), squash (p=0.967) or milk (p=0.184) between active and seated video gaming.

4.3.4 Subjective appetite

There was no detectable difference in the mean baseline appetite sensations of hunger, prospective food consumption or fullness, between any of the four gaming bouts (all p>0.05) (Table 4.4). Differences were detected between the gaming bouts with food and those without food for sensations of hunger, prospective food consumption and fullness. Time-averaged AUC x 120 min revealed the boys felt more hungry during the seated video gaming bout without food compared with when they were both seated (p=0.006) and active video gaming with food (p=0.009) (Table 4.4) (Figure 4.3).

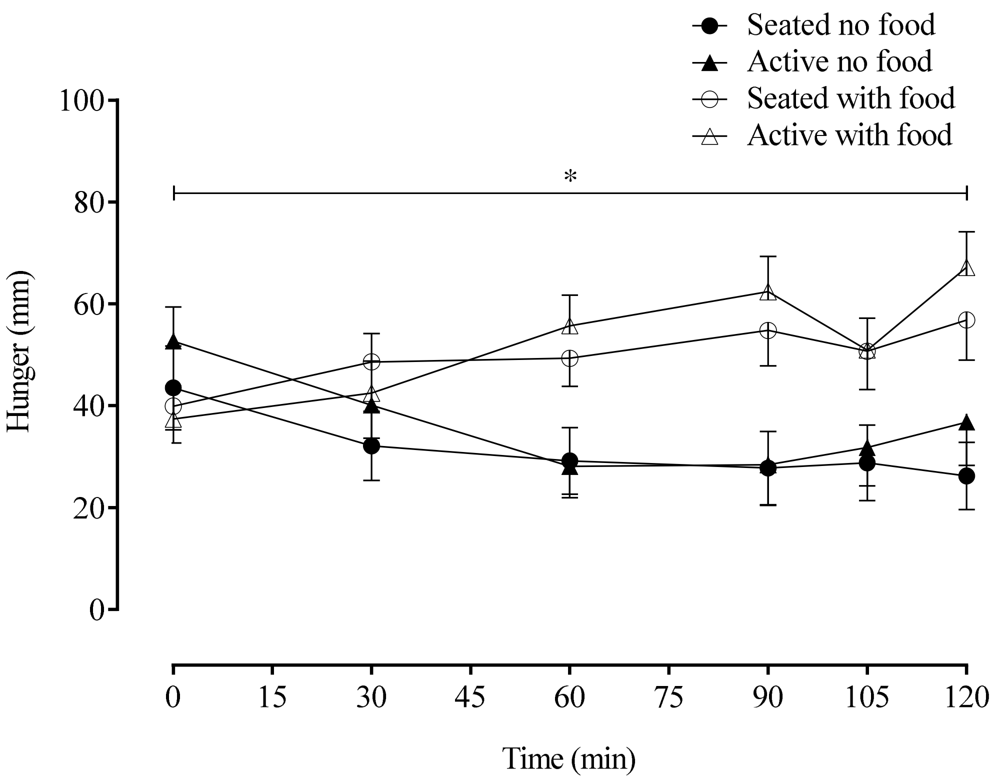

Figure 4.3. Mean $_{\pm SEM}$ hunger sensations. *Time averaged AUC x 120 min revealed during both seated (p=0.006) and active video gaming with food (p=0.009), the boys felt less hungry than during seated video gaming without food.

Table 4.4. Mean $\pm_{SEM}$ values for all baseline and time-averaged AUC appetite sensations of hunger, prospective food consumption and fullness for all four gaming bouts, for all boys (n=21).

		Hunger (mm) (not at all hungry = 100 mm)		Prospective food consumption (mm) (nothing at all = 100 mm)		Fullness (mm) (not full at all = 100 mm)	
	Gaming bout	**Mean**	**SEM**	**Mean**	**SEM**	**Mean**	**SEM**
Baseline	Seated no food	43	8	46	8	55	9
	Active no food	52	7	55	7	60	7
	Seated with food	39	7	41	7	71	6
	Active with food	37	7	38	7	58	8
Time-averaged AUC x 120 min	Seated no food	32	5	32	5	66[c]	5
	Active no food	35	6	38	6	70	6
	Seated with food	48[a]	4	55[b]	6	50[d]	4
	Active with food	50[a]	4	50[b]	5	52[d]	5

[a] During both seated (*p*=0.006) and active video gaming with food (*p*=0.009), the boys felt less hungry than during seated video gaming without food.
[b] During both seated (*p*=0.002) and active video gaming with food (*p*=0.008), the boys wanted to eat more than during seated video gaming without food.
[c] During seated video gaming without food, the boys felt less full than when seated video gaming with food (*p*=0.003).
[d] Indicates during both seated (*p*=0.002) and active video gaming with food (*p*=0.014), the boys felt more full than when active video gaming without food.

In relation to prospective food consumption, the time-averaged AUC x 120 min values showed the boys wanted to eat more when seated video gaming without food than when both seated (*p*=0.002) and active video gaming with food (*p*=0.008) (Table 4.5) (Figure 4.4).

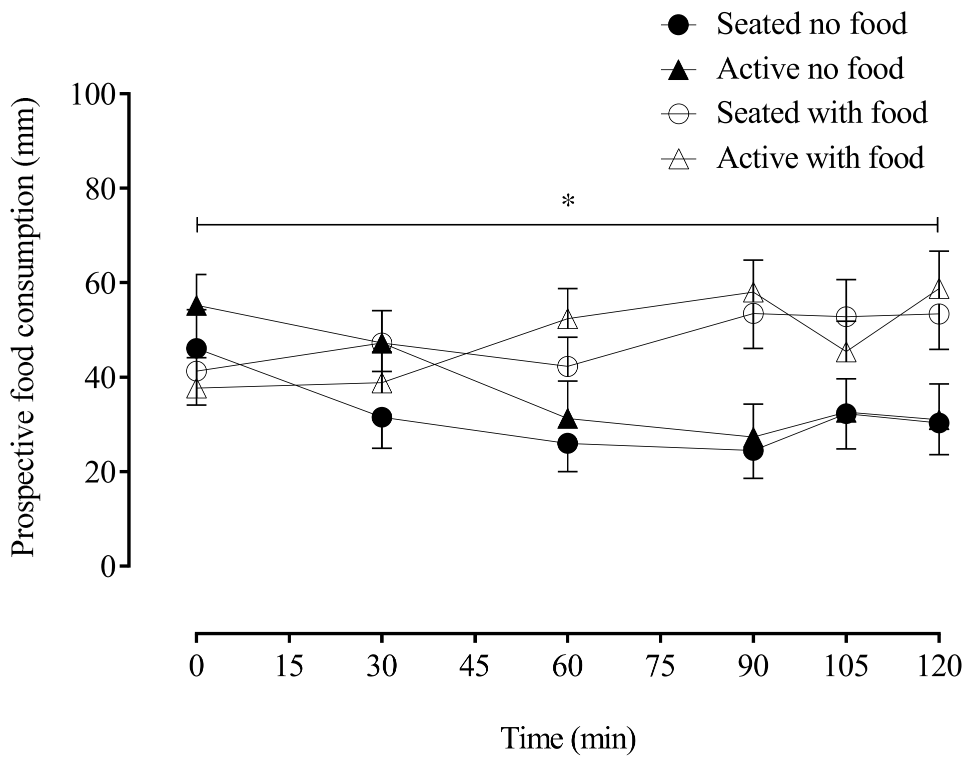

Figure 4.4. Mean ±SEM prospective food consumption for all participants (n = 21). * Time averaged AUC x 120 min revealed the boys wanted to eat more during seated video gaming without food, than during both seated (*p*=0.002) and active video gaming with food (*p*=0.008).

In relation to fullness, the time-averaged AUC x 120 min values showed the boys felt less full during the seated video gaming bout without food than when they were seated video gaming with food (*p*=0.003). The boys also felt less full when active video gaming without food compared with when they were both seated (*p*=0.002) and active video gaming with food (*p*=0.014) (Table 4.5) (Figure 4.5).

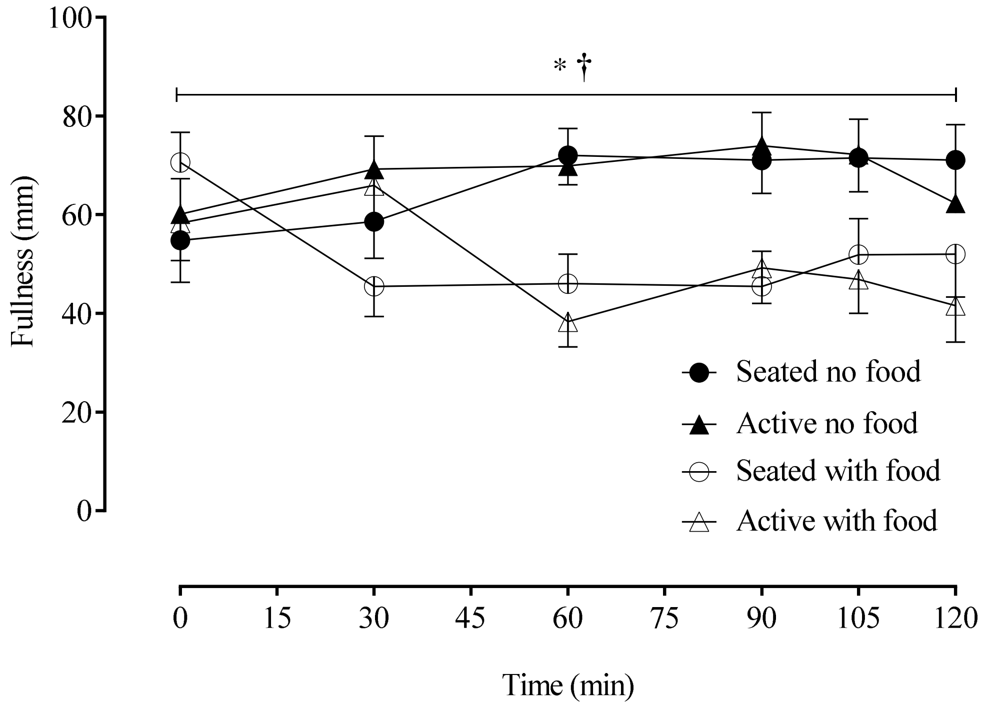

Figure 4.5. Mean ±SEM fullness sensations for all participants (n = 21). *Time averaged AUC x 120 min revealed, during seated video gaming without food, the boys felt less full than when seated video gaming with food (*p*=0.003). †Time averaged AUC x 120 min revealed during both seated (*p*=0.002) and active video gaming with food (*p*=0.014), the boys felt more full than when active video gaming without food.

4.4 Discussion

The present study is the first to have investigated acute *ad-libitum* EI and subjective appetite sensations concurrently, during active video gaming in a paediatric population. The main findings were that no significant differences were apparent in the acute *ad-libitum* EI of 8-11 y boys during 90 min of active video gaming, when compared with seated video gaming. Despite the lack of difference in gaming EI, it took considerably longer for the first eating episode to occur during active video gaming (17.10 min) than during seated video gaming (6.90 min). Furthermore, the greatest proportion of EI (MJ) was from crisps during both gaming bouts and although there were no differences in overall EI, the amount of this particular food consumed, was significantly lower during active than seated video gaming. Physical activity (METS) and subsequent estimated EE were significantly greater during active video gaming than during seated video gaming. Time spent being sedentary was significantly lowered by active video gaming, both with and without food and time spent in light PA was significantly increased during active video gaming without food. During the two bouts with food and drinks offered *ad-libitum*, relative energy balance was significantly lowered due to active video gaming. Differences were also apparent in sensations of hunger, prospective food consumption and fullness but these appeared to be influenced by whether food was made available to the boys during the gaming bouts, rather than by the type of game being played.

To the best of our knowledge, no other studies have compared EI during the play of a genuine active video game with a seated video game in this age group. The lack of difference found in the present study is consistent with EI findings for adults whilst playing both Nintendo Wii™ and Xbox 360 (Lyons et al., 2012). In the cited study, EI following active video gaming was not significantly lower than after seated video gaming. Over the 1 h gaming periods, the adults consumed an average of 3.13±2.26 $MJ \cdot h^{-1}$ when seated, versus 2.32±2.08 $MJ \cdot h^{-1}$ when active. The total energy consumed by the present population was equivalent to 1.92 $MJ \cdot h^{-1}$ during seated video gaming and during active video gaming was 1.53 $MJ \cdot h^{-1}$ and so lower than observed in the adults.

The only comparison that can be made with paediatric gaming EI might be with the study by Mellecker and colleagues (2010) in which 9-13 y old children played a seated video game and an enhanced activity gaming device. In the previous study, the total energy consumed by the children during the seated video gaming bout was equivalent to 1.57 $MJ \cdot h^{-1}$ and during active video gaming was 1.60 $MJ \cdot h^{-1}$. Such values are similar per hour to those found in the present study (60 min seated video gaming 1.92 $MJ \cdot h^{-1}$ versus 60 min active video gaming 1.53 $MJ \cdot h^{-1}$). Although in the present study, EI during seated video gaming was higher and during active video gaming was lower than found by Mellecker and colleagues (Mellecker et al., 2010). Such differences might be due to the former study being conducted in the laboratory and with the active video game being a gaming device played whilst walking on a treadmill. As such the active video gaming format might not have been as stimulating or challenging for the children as an actual active video game. Our intention with the use of Nintendo Wii Sports™ tennis and the primary school settings was to provide a more naturalistic intervention for the participants.

Total EI during 90 min of seated video gaming was calculated to be 2.88 MJ, and for active video gaming 2.30 MJ. When considering the daily estimated average requirement (EAR) for energy is 7.70 MJ for males aged 9 y (the average age of the present study population) in the UK (Jackson, 2011), the EI due to seated and active video gaming equated to 37% and 30% of daily EAR, respectively. Relative energy balance which was calculated for seated and active video gaming by subtracting the value for estimated EE from EI, was found to be 2.42 MJ and 1.64 MJ, respectively. Despite EI being lower during active video gaming with food, it was not offset by the greater amount of energy estimated to have been expended. Instead a substantial energy surplus was produced which could be clinically meaningful in terms of childhood overweight and obesity. It has been suggested that a decrease in dietary intake by 0.46 to 0.69 MJ per day, might be effective in reducing the energy gap and subsequent body mass in children (Wang, Gortmaker, Sobol, & Kuntz, 2006). The energy surplus observed here from a real-life 90 min active video gaming bout, could therefore contribute to a state of positive energy imbalance if not compensated for later, by a down-regulation in EI or up-regulation in EE.

In addition, when the individual foods (crisps and apple) and fluids (fruit squash and semi-skimmed milk) consumed by the boys during gaming were examined it was revealed that 59% (1.70 MJ) of seated and 54% (1.25 MJ) of active gaming EI was from crisps. Therefore, at cessation of the gaming bouts, the greatest contributor to the positive, relative energy balance state of the boys was crisps, an energy dense food of low micronutrient value. When considering the aforementioned proposed reduction in daily dietary intake (Wang et al., 2006) and accounting for the significantly lower levels of EI from crisps alone, during active video gaming, if consumed frequently during both active and seated video gaming, this particular food has the potential to contribute to a positive energy balance state.

Due to this present study being an acute investigation, the EI of the boys was not monitored after the gaming bouts had ended (17:00) so it is not known whether compensation for the gaming EI occurred later. However, during seated video gaming with 15-19 y adolescents, EI was monitored for the remainder of the day. Nonetheless compensation did not occur for the extra food they had consumed during the seated bout up to this time-point (Chaput et al., 2011). Although an exercise induced increase in EE previously implemented in 13-15 y old well trained girls, observed that compensation for the extra energy expended did not occur until 72 h later (Rumbold et al., 2011). Future research that examines compensation post active video gaming is warranted.

In relation to subjective appetite, no significant differences were detected in sensations of hunger, prospective food consumption or fullness between the seated and active video gaming bouts in which foods were available *ad-libitum*. Unsurprisingly, the boys reported feeling more hungry and wanted to eat more when both seated and active video gaming without food, in comparison to the corresponding bouts with food. Consequently, they felt more full when both seated and active video gaming with food. To the best of our knowledge, no other paediatric studies have examined subjective appetite responses to active video gaming so there are no studies with which these results may be directly compared. With regards to seated video gaming there have been two previous paediatric appetite studies that have compared the acute appetitive effects of seated video gaming with resting. Both measured and compared the acute appetitive effects of seated video gaming versus resting. In 11-13 y boys, EI was found to be significantly lowered by 0.25 MJ after 30 min of seated video gaming (Branton et al., 2014). Whilst 15 to 19 y adolescent males consumed 0.09 MJ more, after one h of seated video gaming but this was not significantly higher than EI after resting (Chaput et al., 2011). It is important to note that in the more recent cited study, a glucose pre-load was administered at the start of the session which might have suppressed subsequent food intake (Branton et al., 2014).

The lack of difference in sensations of hunger, prospective food consumption and fullness and EI between seated and active video gaming with food in the present study population, and during seated video gaming in 15-19 y adolescents might not be entirely coincidental. It is possible, that both seated and active video gaming could lead to over-consumption of energy without an increase in hunger, as previously reported with television watching (Temple et al., 2007). When supplied with a 1000 kcal portion of a favourite snack food whilst watching an unbroken television programme for 23 min, 9-12 y-old children consumed more and spent longer eating (min), than a no television control group and a group that watched 2.5 min clips repeatedly. It is thought the distractive effect of television disrupts habituation to food cues that are controlled by assimilated signals from the sensory, neuronal and digestive systems (Temple et al., 2007). The disruption in habituation to food cues has been labelled as the mental-stress-induced reward system. A consequence of the mental-stress-induced reward system is that homeostatic appetite-related signals appear to go unheeded causing over-compensation in EI for the energy being expended (Chaput et al., 2011; Dallman, 2010; Marsh et al., 2013). In relation to active and seated video gaming, this disruption, coupled with an environment in which highly palatable foods are available *ad-libitum*, could have activated the brain's reward centre. As a result, the effects of the satiety peptides might have become suppressed by an augmented release of hormones associated with pleasure (dopamine, endocannabinoids and opiates) could have occurred. This response sustains the drive to eat via an interaction between hedonic and homeostatic systems (Finlayson, King, & Blundell, 2008). Further research which measures satiety hormones objectively alongside subjective appetite sensations and EI is warranted within the present paediatric population, to investigate whether these signals do indeed become suppressed during active and seated video game play and whether there are differences between the two gaming bouts.

In the present study, PA METS were greater due to active video gaming in comparison to seated video gaming which is in agreement with previous acute paediatric active video gaming studies that have also utilised Nintendo Wii™ Sports tennis (White et al., 2011). When the boys were gaming without food, mean PA intensity when seated was established as sedentary (1.38±0.07 METS), and when playing Nintendo Wii™ Sports tennis was light intensity (2.14±0.10 METS) indicating an increase in PA METS by 55%. In the gaming bouts with food, the boys maintained similar PA METS to the gaming bouts without food. When seated video gaming with food, the mean PA intensity was sedentary (1.49±0.07) METS) and active video gaming was light intensity (2.08±0.09) MET). Former studies have also recorded similar PA intensities with 11-17 y children achieving 2.40 METS (Graves et al., 2008) and 11 y boys attaining 2.16 METS (White et al., 2011) whilst playing Nintendo Wii™ Sports tennis. The PA METS, although slightly higher than established in the present study were likely due to the much shorter laboratory-based bouts of gaming used in the former studies and greater maturity and thus physical strength of the participants (Graves et al., 2008; White et al., 2011).

Playing a real-life bout of Nintendo Wii™ Sports tennis (Allsop et al., 2013) however, did not achieve the desired moderate to vigorous levels of PA, recommended for UK children for at least 60 min per day (Almond et al., 2011). The mean maximum time the boys spent in MVPA was only 1.57 min, during the active video gaming bout without food. Real-life active video gaming cannot therefore be considered as a complete replacement for other exercise or sports that meet with MVPA. In spite of this, active video gaming both with and without food produced a decrease in time spent being sedentary (active gaming without food 22.38 min; active gaming with food 25.48 min) compared to seated gaming both with food (55.29 min) and without food (64.29 min). During the 90 min of active video game play, the boys spent the majority of time in light PA (without food 66.05 min; with food 63.33 min) which was significantly greater than when they were seated gaming (without food 25.33 min; with food 34.67 min). The shift from sedentary to light PA that was observed during active video gaming, is noteworthy particularly in view of the specific guideline set by the Chief Medical Officers for the UK, for 5-18 y children to minimise time spent being sedentary (Almond et al., 2011). Essentially, active video game play has the potential to be used as a supplement for PA to help children reduce their sedentary time. For this reason, active video gaming should be encouraged over seated video gaming. There should be further work however to determine the effects on children's sedentary behaviour and subsequent health outcomes.

To the best of our knowledge, this is the first paediatric study to investigate the influences of active video gaming on acute subjective appetite sensations and EI. The strengths of the study were that it utilised an intervention designed from actual survey findings that established the real-life active video gaming practices of 7-11 y children. In addition, the gaming bouts were implemented as an after-school club thus creating a more relaxed and naturalistic setting for participants, in contrast to that of a laboratory. Such a free-living and holistic design has evolved from earlier paediatric exercise and appetite research (Dodd et al., 2008; Rumbold et al., 2011; Rumbold et al., 2013).

A limitation of the study was that only Nintendo Wii™ Sports tennis was utilised during the two, 90 min active video gaming sessions, however, it was important for the boys to play the same game to enable accurate comparison of the individual gaming bouts. Furthermore, the Nintendo Wii™ Sports tennis game utilises both upper and lower limbs during play, which should allow for greater body movement and thus higher activity counts and EE (Graves et al., 2008; Maddison et al., 2007). It is also pertinent to acknowledge that the prediction of EE from METS obtained by accelerometry is not without error, particularly in children (Nilsson et al., 2008; Ridley, Ainsworth, & Olds, 2008). It was considered more important however to implement the active video gaming sessions in a manner that was most true-to-life than would have been, if conducted in the laboratory. For this, it was necessary to measure PA by accelerometry and thus estimate EE and relative energy balance. Consequently, both estimations will not be without error, but then all methods which provide a measure of EE have some degree of inaccuracy. The advantage of utilising this method was that it enabled the present study to maintain an exploratory approach. Future work should consider investigating the reliability of active video gaming EE estimations by accelerometry and use the paediatric responses to active video gaming presented in this study to help power future studies.

Due to the novel design of the present study, a power calculation to determine sample size could not be carried out prior to data collection. A retrospective power calculation based on the results obtained for relative energy balance was thus performed. The retrospective power calculation established that a sample size of at least 17 children would provide statistical power of 80%. The present study recruited a sample size of 21 boys, thus demonstrating that the statistically significant results obtained were adequately powered.

The findings of the present study suggest that the availability of food had a significant effect on sensations of hunger, prospective food consumption and fullness. However, during game play with food, there were no differences in the acute appetite sensations of the 8-11 y boys between active and seated video gaming. Subsequent EI during seated and active video gaming was considerable as this was estimated to be 37% and 28% respectively, of total daily EAR (based on 9 y old boys), with energy from crisps making the greatest contribution. Despite this, active video gaming was found to contribute to PA recommendations by increasing PA levels from sedentary when seated video gaming, to light PA. Consequently, at the end of the active video gaming bout with food, the relative energy balance was estimated to be 0.87 MJ lower (1.50 MJ) than for seated video gaming (2.37 MJ). Nonetheless, if the REI for both gaming bouts was not compensated for later it could contribute to a positive energy balance state.

When considering the potential risk that EI during active and seated video gaming might have to a child's energy balance, a rational step for further research would be to explore the homeostatic, appetitive mechanisms related to satiety in an effort to explain this eating behaviour. It is important to understand whether children's satiety signals are suppressed whilst active video gaming and so further work should seek to determine the levels of the circulating satiety-related signals, $GLP\text{-}1_{7\text{-}36}$, leptin, glucagon, insulin and glucose, in 8-11 y old boys. However, the day-to-day variability of these satiety-related signals must be an important consideration. Therefore, firstly the day-to-day variation of $GLP\text{-}1_{7\text{-}36}$, leptin, glucagon, insulin and glucose needs to be established in this population group in a fasted state. In doing this, a similar active video gaming design to that used in the present study could be utilised to determine whether the satiety signals of 8-11 y old boys are indeed suppressed by active video game play. Acute subjective appetite (hunger, prospective food consumption and fullness) and EI responses to active video gaming also require further exploration, in order to establish whether the observed acute over-compensation in EI is offset by subsequent EI and EE, after a gaming session has ended.

CHAPTER V

SATIETY-RELATED SIGNALLING, SUBJECTIVE APPETITE AND ENERGY INTAKE, IN RESPONSE TO ACTIVE AND SEATED VIDEO GAMING, IN 8 TO 11 YEAR-OLD BOYS

5.1 Introduction

Active video gaming can elicit light to moderate intensity PA in children, as identified in Chapter IV and by former studies (Graves et al., 2008; White et al., 2011). However, the PA level attained appears to be dependent on the gaming console, the active video game and the time spent playing. In the laboratory, Nintendo Wii™ and Nintendo Wii™ Sports have been found to elicit two to three times more energy and between 2 and 6 METS in paediatric populations (Graf et al., 2009; Graves et al., 2008; Mitre et al., 2011; White et al., 2011). Yet in a more naturalistic setting and utilising a gaming protocol that was representative of the true-to-life active video gaming practices of 7-11 y children, the PA levels were established to be only light intensity exercise (2 METS) (Allsop, Dodd-Reynolds, Green, Debuse & Rumbold, 2015). In terms of health therefore, it seems more likely that real-life active video gaming could contribute to a reduction in the time children spend being sedentary, rather than supplementing MVPA although further research is necessary.

In relation to sedentary screen based media, there is evidence from studies with children of a reduction in PA, not only due to television viewing but also from computer use and seated video gaming (Atkin et al., 2013; Hands et al., 2011; Huang et al., 2013). In addition, findings suggest that television viewing and computer use can induce unhealthy food intake (Berentzen et al., 2014; Pearson & Biddle, 2011). Consequently, sedentary screen based media activities could contribute to a positive energy balance state (Hands et al., 2011; Vandewater et al., 2004).

Findings from studies that have explored the effects of seated video gaming on subjective appetite sensations and EI concurrently, in paediatric groups, thus far are ambiguous (Branton et al., 2014; Chaput et al., 2011; Marsh et al., 2013). Two of the cited studies compared sensations of hunger, prospective food consumption and fullness and subsequent EI from an *ad-libitum* test meal, following seated video gaming and resting in 11-13 y boys (Branton et al., 2014) and 15-19 y adolescent males (Chaput et al., 2011). The EI of 11-13 y boys was lower following seated video gaming and the corresponding subjective appetite sensations demonstrated reduced hunger and prospective food consumption and increased fullness throughout the bout (Branton et al., 2014). Yet in 15-19 y adolescent males, EI was higher following seated video gaming but there were no significant differences in appetite sensations, between seated video gaming and resting (Chaput et al., 2011). The more recent of these former studies provided glucose and sucralose pre-loads at the beginning of the bouts however and this led to decreased sensations of hunger, prospective food consumption and increased fullness and was the likely reason for the subsequent reduction in EI in the boys (Branton et al., 2014).

Marsh and colleagues (2013) developed the seated video gaming research further by comparing this activity with television viewing and recreational computer use and by offering food and drinks *ad-libitum* throughout the individual bouts. During seated video gaming and television viewing trials, the average appetite score (Bellissimo et al., 2007) of the 9-13 y boys decreased significantly in comparison to the computer trial, and this led to lower EI in the latter mentioned trial (2.87 MJ). There were no significant differences in total acute *ad-libitum* EI between seated video gaming and television viewing and considerable amounts of food and drink were consumed during these trials (television viewing 3.44 MJ and seated video gaming 2.91 MJ) (Marsh et al., 2013). The results of studies by Marsh et al., (2013) and Chaput and colleagues (2011), provided evidence that during and following seated video gaming there can be substantial EI. Furthermore, the EI was not compensated for at a later time-point during the day in the adolescents, either by a down-regulation of EI or an up-regulation of EE (Chaput et al., 2011). In the 9-13 y boys, compensation was not monitored beyond the laboratory trials (Marsh et al., 2013).

More recently, EI and subjective appetite in response to individual 1 h bouts of active video gaming, seated video gaming and resting were examined in adolescent males (Gribbon et al., 2015). In the cited study, EE was significantly greater during active video gaming than during the seated video gaming and resting conditions. When EI and physical activity energy expenditure (PAEE) were monitored over 24 h it was established that compensation for the extra EE occurred later through an up-regulation in EI. As a consequence, 24 h following all three conditions, the adolescents were found to be in a similar positive energy balance state (Gribbon et al., 2015). However, both Gribbon and colleagues (2015) and Chaput et al., (2011) did not offer food or drinks *ad-libitum* during the active video gaming but following the seated video gaming and resting conditions. In relation to active video gaming this might not be representative of real-life practices since EI was reported to occur during game play by 7-11 y children (Allsop et al., 2013) and more recently by adolescents (Simons et al., 2015). Chapter IV of this thesis has also provided evidence of significant EI during both seated and active video gaming in 8-11 y boys, which during the latter bout was not compensated for by the energy expended during game play (Allsop et al., 2015). Similar to 9-13 y boys (Marsh et al., 2013), EI and EE were not monitored beyond the active and seated video gaming bouts. Further active video gaming appetite and EI research is required therefore, to investigate whether compensation for the extra EE and *ad-libitum* EI during game play occurs at a later time-point.

It has been proposed that the distractive effect of television viewing could disrupt the response to food cues, a mechanism known as dishabituation (Temple et al., 2007). There also appears to be evidence of the mental-stress-induced reward system from seated video gaming (Chaput et al., 2011) which is a response that overrides homeostatic fullness cues and sustains the drive to eat and is akin to the hedonics of food intake (Finlayson et al., 2008; Yeomans, 1998). In addition to subjective measures, Chaput and colleagues (2011) measured the concentrations of the homeostatic hunger-related signals, total ghrelin, serum insulin and plasma glucose, during seated video gaming and resting. No differences were found in the concentrations of total ghrelin or serum insulin and therefore, no evidence of increased hunger during or following seated video gaming and resting. Yet blood glucose was significantly raised during seated video gaming and according to the 'glucostatic theory' of short-term appetite regulation, this is indicative of a satiety response (Mayer, 1952). Despite the increased concentrations of glucose during seated video gaming however, the adolescents consumed more in the post-gaming meal and subjective sensations of hunger, prospective food consumption and fullness were no different to those measured during resting (Chaput et al., 2011). The proposed satiety response thus appeared to be overridden by hedonic mechanisms causing an over-compensation in EI, for the energy expended (Finlayson et al., 2008; Yeomans, 1998). Satiety-related hormones were not measured by Chaput and colleagues (2011) in this earlier seated video gaming study and so it could not be confirmed if there was indeed a satiety response.

In Chapter IV, the acute appetite sensations and EI of 8-11 y boys were compared during two 90 min bouts of active video gaming with two, 90 min bouts of seated video gaming. When foods and drinks were offered *ad-libitum* during one of the active video gaming bouts and one of the seated video gaming bouts, the EI of the boys was not significantly different. Indeed the acute EI due to eating and drinking during the active video gaming bout was established as 27% (2.30±0.28 MJ) of daily EAR (based on 9 y old boys). Subsequently, the relative energy balance was estimated to be 1.64±0.28 MJ and it was not established whether this acute over-compensation in EI was offset by any subsequent decrease in EI, or increase in EE following the gaming bout. Furthermore, there were no differences between seated and active video gaming with food in the acute subjective appetite sensations of hunger, prospective food consumption and fullness. A more in-depth understanding of the acute appetite responses of 8-11 y-old boys is therefore required, to try to establish whether the acute EI in response to active video gaming is more likely due to homeostatic or hedonic mechanisms.

Two of the same gaming bouts utilised in Chapter IV, which were a 90 min active video gaming bout and a 90 min seated video gaming bout during which foods and drinks were offered *ad-libitum* will be used in the present study. The primary aims of this study were to explore the acute satiety-related signalling of plasma GLP-$1_{7\text{-}36}$ and blood glucose, subjective appetite and EI of 8-11 y boys in response to both active and seated video gaming. The study will therefore measure the acute responses of plasma GLP-$1_{7\text{-}36}$ and blood glucose during and following 90 min bouts of active and seated video gaming. Alongside the objective measurement of appetite, the subjective sensations of hunger, prospective food consumption and fullness will also be examined. In addition, the EI of the boys will be measured during the gaming bouts and 1 h following them, in a test meal.

5.2 Materials and methods

5.2.1 Preliminary reliability testing of satiety-related signals

In human appetite regulation, GLP-$1_{7\text{-}36}$, glucagon, leptin and insulin represent some of the more commonly studied hormones in research and clinical practice that are known to play a major role in satiety signalling (Gautron & Elmquist, 2011; Jiang & Zhang, 2003; Morton, Cummings, Baskin, Barsh, & Schwartz, 2006; Williams et al., 2009). The aforementioned hormones are typically sampled by the antecubital-venous method. When considering the young age and thus vulnerability of the present population, this sampling method was deemed too invasive and may have even been regarded as unethical. Recently, a study with healthy adults was conducted which examined the agreement between plasma GLP-$1_{7\text{-}36}$, glucagon, leptin and insulin obtained from fingertip capillary samples, with those drawn by antecubital-venous sampling (Green et al., 2014). Green and colleagues (2014) established that fingertip capillary blood sampling was an appropriate and reproducible method for quantifying GLP-$1_{7\text{-}36}$ and glucagon. Such a method is far less invasive than venous sampling, thus a more suitable procedure for use in paediatric populations (Green et al., 2014). As a preliminary investigation therefore, the extent of between-day variation and the reproducibility of the aforementioned fingertip-derived satiety-related peptides and glucose were measured, in 8-11 y boys. The methods and materials utilised are presented in full in Appendix J.

5.2.2 Study design

A randomised, cross-over design was used to compare the acute satiety-related signals, subjective appetite and EI responses of 8-11 y-old boys, to two gaming bouts each separated by 1 week, utilising methods identified in Chapter IV. The two gaming bouts were: 1) 90 min of seated video gaming with food and drinks offered *ad-libitum* and 2) 90 min of active video gaming with food and drinks offered *ad-libitum*. The boys were stratified according to school year into groups of two. Each group was then randomly assigned to a different bout every week so that by the end of the 2 weeks they had completed each trial.

The study was approved by the University of Northumbria, Faculty of Health and Life Sciences Ethics Committee. Written informed consent was obtained from each child's parent (or main carer) and assent from every boy, prior to data collection.

5.2.3 Recruitment

The same recruitment procedure was utilised as in Chapter IV which is described in full in section 4.2.2. To enable recruitment of 8-11 y boys, the Head Teacher of a primary school within the city of Newcastle upon Tyne (North East England, UK) was approached by letter, sent by email. Following a response from the Head Teacher, a further email was sent to arrange a meeting to explain the study in greater detail. During this meeting the Head Teacher consented to the recruitment and participation of male pupils aged 8-11 y. During the recruitment process, a total of 60 recruitment packs were distributed to eligible boys who expressed an interest in taking part in the study.

To determine the sample size for the study, a power calculation was carried out based on the relative energy balance of the 8-11 y boys observed in Chapter IV, section 4.3.3. The power calculation established that a sample size of at least 17 children would provide statistical power of 80%. Signed consent forms were received from 22 boys, although one boy was unable to provide blood samples during the trials, all other data collected from him was included in the analysis. It was necessary to remove data for one boy as his food diary and dietary recall showed non-standardization of EI and so 21 boys took part overall.

5.2.4 Anthropometric assessment and familiarisation

Prior to the first gaming trial, each boy was anthropometrically assessed and familiarised with the gaming consoles and games and research materials used as described in section 4.2.3.

5.2.5 Study protocol

Prior to first gaming intervention, preliminary blood samples were taken for each boy to establish fasted levels of GLP-$1_{7\text{-}36}$ and glucose, the full procedure, methods, materials and results of this testing are provided in Appendix J. Following the preliminary blood sampling and before the first gaming intervention, each boy was provided with a self-report, weighed food diary and a set of food weighing scales (Salter©, Kent, UK). With the help of their parent they were asked to weigh and record all foods and drinks consumed from 17:00 the evening before each intervention day, until breakfast on the morning of each intervention day. Following the first gaming trial, a copy of the food diary was provided to each boy and aided by their parent (Bates et al., 2014) they were asked to replicate their food and drink intake prior to the second intervention day, 1 week later. The boys were also asked to abstain from all PA from 17:00 the evening before, until the morning of each intervention day.

5.2.5.1 Gaming interventions

The design of the gaming protocol was based on Chapter III which described the real-life active video gaming practices of 7-11 y children from Newcastle upon Tyne (UK) (Allsop et al., 2013). The gaming console used for the active video gaming bouts was Nintendo Wii™ and the game used was Nintendo Wii™ Sports tennis (Allsop, et al., 2013). The seated video game utilised was 'Mario and Sonic at the London 2012 Olympic Games' which was played on the handheld device, Nintendo© 3DS. The two gaming interventions were separated by 7 days and took place on the same school day of each week over two consecutive weeks. As in Chapter IV, the two gaming bouts were: 1) 90 min seated video gaming with food and drinks offered *ad-libitum*; 2) 90 min active video gaming with food and drink offered *ad-libitum*. The gaming bouts were implemented in the morning in the University laboratory. The boys were tested in groups of two and randomised to both play the same gaming console and game. When assigned to the Nintendo Wii™, the boys played together against the computer, and when they played the Nintendo 3DS they played individually against the computer. In doing this, peer influence related to winning or losing was avoided along with any subsequent effects on EI. As far as possible, the boys were paired according to year group. The boys were randomly assigned to active or seated video gaming so that by the end of 2 weeks they had completed each trial and acted as their own controls.

On the intervention days, the boys were met at school at 0830 h by two members of the research team and transported to the University laboratory. On arrival (0850 h), they were able to relax until 0900h when they completed the VAS for hunger, prospective food consumption and fullness. Following this, a fingertip blood sample was taken from each boy to enable the determination of baseline plasma GLP-$1_{7\text{-}36}$ and blood glucose. An accelerometer (Actigraph LLC© GT3X+) which had been programmed to record PA counts in 10 s epochs was then placed on the right hip of each boy. The foods and drinks were placed at a station designated to each of them and they were asked to commence gaming for 90 min. The boys completed appetite VAS at 45 min during gaming, at the end of gaming (90 min) and 45 min post gaming and were also taken immediately following the test meal. At the same time points up to the test meal, further fingertip blood samples were taken for the determination of plasma GLP-$1_{7\text{-}36}$ and blood glucose. Upon termination of each 90 min gaming bout, the participants rested for 60 min following which they were provided with an *ad-libitum* test meal, before being returned to school by the research team (Figure 5.1).

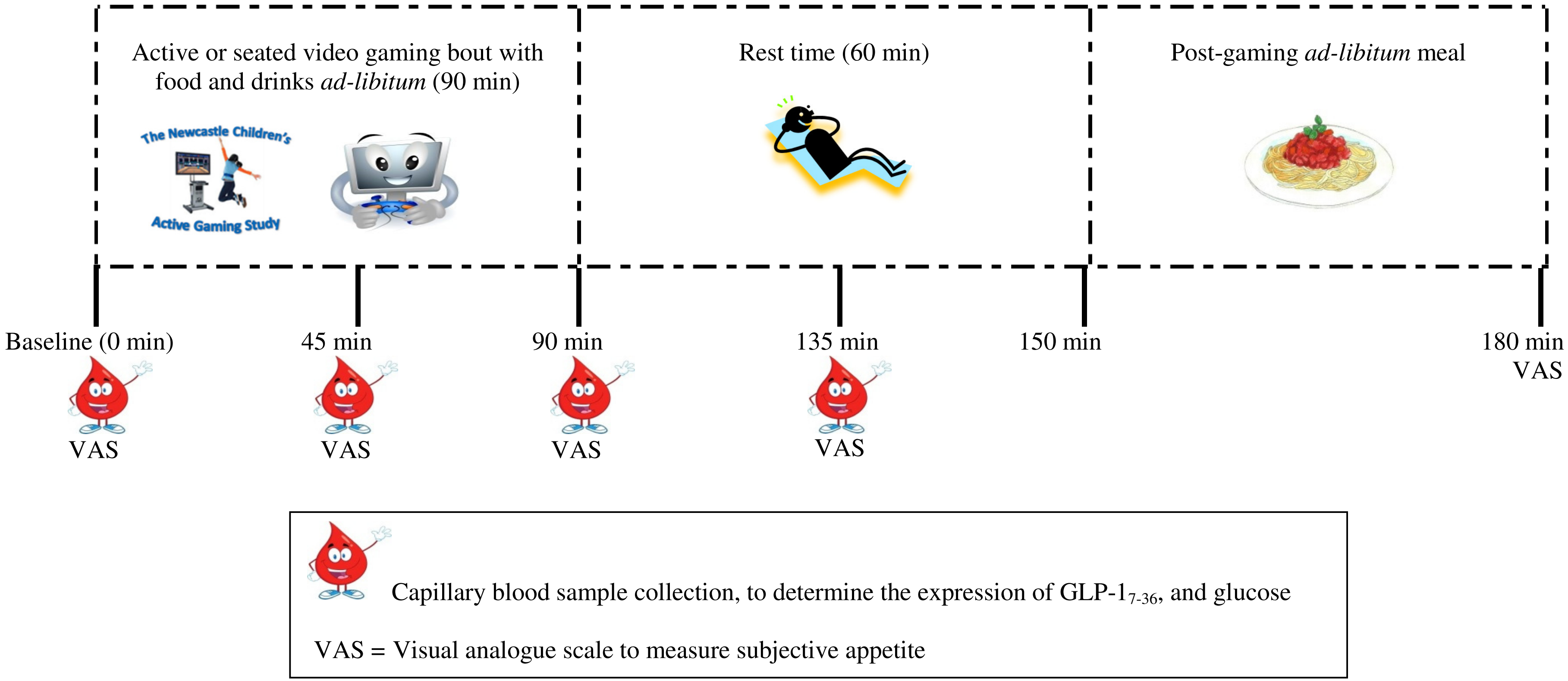

Figure 5.1. Schematic representation of gaming trials

5.2.6 Physical activity assessment and energy expenditure

During both 90 min gaming bouts, the PA levels of each boy, the mean time (min) spent sedentary, in light PA and MVPA, were measured and EE was estimated, as detailed in Section 4.2.5.

5.2.7 Energy intake and relative energy balance

The food and drink items provided *ad-libitum* during the 90 min gaming bouts were based on previous findings (Allsop et al., 2013) and are displayed with respective macronutrient values in Table 5.1. The food and drink items were provided to the boys as in Chapter IV and described in section 4.2.6. When the gaming bouts commenced, the first eating episode for each boy was timed (min) and recorded.

Table 5.1. Serving size, total energy and macronutrient values of food and drink items served during the gaming bouts.

Food or drink	Serving size	Energy (MJ)	Carbohydrate (g)	Fat (g)	Protein (g)
Apples ("Royal Gala" raw, sliced and cored)	130 g	0.26	15.60	0.13	0.52
Walker's© ready salted crisps	50 g	1.10	25.75	15.95	3.05
Semi-skimmed milk	250 mL	0.52	12.00	4.50	9.00
"Jucee" apple and blackcurrant squash (no added sugar)	250 mL (1:5 dilution)	0.03	2.50	0.00	1.30

The *ad-libitum* test meal provided was pasta, with tomato sauce, cheddar cheese and olive oil (ASDA, Leeds, UK) which was served in excess. They were instructed to eat until they felt comfortably full, at which point the meal was terminated. As they ate the test meal, the bowl was refilled by the researcher. The total time for eating was measured (min) and recorded for each boy. The research team covertly weighed the test meal before it was served and as the meal was terminated. The macronutrient content of the meal was 58% CHO, 28% fat and 14% protein (PRO) and provided 450 kJ (107.5 kcal) per 100 g of total energy, similar to a pasta meal used in a previous adolescent appetite study (Rumbold et al., 2013). Energy intake for all of the food and drink items served was estimated from individual food labels, an online resource (www.asda.com) and MicroDiet (Downlee Systems©, Derbyshire, UK).

Following the two trials, EE was subtracted from the amount of energy consumed during each 90 min gaming bout, for each boy. Relative energy balance was calculated after each 90 min gaming bout as follows;

Gaming EI (MJ) – Gaming EE (MJ).

Relative energy was calculated to include EI of the test meal as follows;

Gaming EI (MJ) + test meal EI (MJ) – gaming EE (MJ).

5.2.8 Subjective appetite

Appetite sensations were assessed using VAS as in Chapter IV and described in section 4.2.7. The boys were requested to rate their sensations of hunger, prospective food consumption and fullness at set times, prior to, during and following gaming on both intervention days. Scales were collected prior to the commencement of gaming (baseline t=0 min), at 45 min during, at the end of gaming (90 min), at 45 min (15 min before test meal) and 90 min post-gaming (after test meal).

5.2.9 Blood sampling

For the measurement of $GLP\text{-}1_{7\text{-}36}$, capillary blood samples were collected immediately before the gaming bouts commenced (baseline t=0 min), midway during the gaming bout (45 min), at the end of the gaming bout (90 min) and at 45 min post-gaming (135 min) (Figure 6.1).Before drawing the sample, each boy pre-warmed the sampling hand in warm water to promote adequate blood flow. The hand was dried thoroughly and the appropriate puncture site was cleaned with an aseptic wipe. Participants were seated upright to control for any postural related changes in plasma volume. The fingertip was pierced with a sterile automated lancet (Accu-Check, Mannheim, Germany) and the blood collected into a pre-cooled EDTA microvette. Prior to blood collection, 33 µL per mL of aprotinin and 30 µL per mL of DPP-IV inhibitor were added to each microvette to prevent the cleavage of GLP-$1_{7\text{-}36}$ by proteases and to aid in its preservation. Immediately following blood collection, the microvettes were placed on ice and then spun at 1500 *g* for 10 min in a multispeed microcentrifuge. Aliquots of the plasma supernatant were pipetted into labelled Eppendorfs and stored at −80 °C for the quantification of GLP-$1_{7\text{-}36}$ at a later time-point.

At the same time as blood was collected for the determination of GLP-$1_{7\text{-}36}$, a sample was also obtained from the same puncture site to establish blood glucose concentrations. Each of these blood samples (0.02 mL) was drawn into a sodium heparinised capillary tube and transferred into an Eppendorf containing 1 mL haemolysis solution (EKF Diagnostics). Samples were shaken to encourage haemolysis and immediately placed on ice.

5.2.10 Blood analysis

The levels of active GLP-$1_{7\text{-}36}$ were simultaneously quantified by electrochemiluminescence, utilising a human hormone multiplex assay (Sector Imager 2400, MesoScale Discovery, Rockville, MD, USA). A summary that describes how the assay detects and quantifies GLP-$1_{7\text{-}36}$ and a detailed copy of the materials and methods used, is provided in Appendix K. To reduce inter-assay variation, samples from each participant were analysed on the same assay plate where possible. Two assay plates were required to analyse all plasma samples collected. The intra-assay CV's were established by the repeated measure of one baseline fingertip capillary plasma sample, due to the small amount of plasma available from the participant samples. For plate one, the CV for GLP-$1_{7\text{-}36}$ was 4% and for plate two was 7%.

The blood glucose samples were quantified by the glucose oxidase method using an automated glucose analyser (Biosen C line, EKF Diagnostics). The method electro-chemically measures β-D-glucose as it is converted to gluconic acid. Prior to use, the analyser was calibrated with a solution of known glucose concentration (12 mmol/L).

5.2.11 Statistical analysis

IBM© SPSS (version 22.0, SPSS Inc., Chicago, Illinois) was used for all statistical analyses. Means ±SEM are presented for all data which was checked for normality using Shapiro-Wilk. When data was not normally distributed, log transformation was performed and outliers removed if necessary. Data was included from 20 boys for GLP-$1_{7\text{-}36}$ (pg/mL) and blood glucose The responses of plasma GLP-$1_{7\text{-}36}$ (pg/mL) and blood glucose (mmol/L), were calculated as time-averaged area under the curve AUC x 135 min for both gaming trials. In addition, plasma GLP-$1_{7\text{-}36}$ and blood glucose responses were calculated as time-averaged area under the curve AUC x 90 min, to establish whether there was a specific effect of the gaming bout. The VAS ratings (mm) for subjective hunger, prospective food consumption and fullness were calculated as time-averaged AUC x 180 min for both gaming trials. They were also time-averaged AUC x 90 to determine if there was a specific effect of the gaming bout. Time-averaged AUC x 135 min and AUC x 90 min values for plasma GLP-$1_{7\text{-}36}$ and glucose, time-averaged AUC x 180 min and x 90 min for subjective sensations of hunger, prospective food consumption and fullness, along with gaming and test meal EI, macronutrient intake (CHO, fat and PRO), the individual foods and fluids consumed during gaming, PA (METS), time spent (min) in sedentary, light and MVPA, estimated EE (MJ), relative energy balance (MJ), time to eating onset during gaming (min) and the ingestion time of the test meal (min), were analysed using paired t-tests. When significant differences were found in the t-test analyses, Cohen's *d* effect size was calculated and interpreted against the effect size categories of ≤0.20 = small effect, ~ 0.50 = moderate effect, and ≥0.80 = large effect (Cohen, 1992). Statistical significance was set at $p< 0.05$ for all analyses.

5.3 Results

5.3.1 Preliminary reliability testing of satiety-related signals

The results of the preliminary reliability testing of the satiety-related signals, GLP-$1_{7\text{-}36}$, glucagon, leptin, insulin and glucose are provided in Appendix J. In brief, the between-day variation of only plasma GLP-$1_{7\text{-}36}$ and glucose were low, with values below pre-determined clinically meaningful differences [0.2 pg/mL (Verdich et al., 2001) and 0.5 (Nair et al., 2009) for GLP-$1_{7\text{-}36}$ and glucose, respectively]. In addition, both plasma GLP-$1_{7\text{-}36}$ and glucose showed good reproducibility. The preliminary study demonstrated that GLP-$1_{7\text{-}36}$ and glucose could be confidently measured by the fingertip capillary sampling method, in 8-11 y boys (Appendix J).

5.3.2 Population Characteristics

The mean age of the boys was 9.9±0.2 y, mean $_{\pm SEM}$ stature 1.45±0.02 m, body mass 37.9±1.6 kg, waist circumference 64.3±1.7 cm and BMI 18.1±0.7 kg/m^2. According to UK age and sex-specific BMI centiles (Cole et al., 1995), the majority of the boys were classified as having a healthy body mass (77.3%), 9.1% were overweight and 13.6% as obese. The mean maturity offset was -3.3±0.1y indicating that as a group the boys were an average of 3 y and 1.2 months from peak height velocity and were of similar maturation status. In addition, all boys were identified as being unrestrained eaters according to the Dutch Eating Behaviour Questionnaire for children (van Strien & Oosterveld, 2008) with a mean $_{\pm SEM}$ dietary restraint score of 1.8± 0.13 categorized as being average for boys of this age (1.53±0.06).

5.3.3 Physical activity levels and energy expenditure

Active video gaming elicited only light PA and seated video gaming was sedentary (Table 5.2). The PA level (METS) during active video gaming were significantly greater than seated video gaming (p<0.001, moderate effect size 0.7). When EE (MJ) was estimated from PA, active video gaming elicited significantly greater energy than seated video gaming (p<0.001, moderate effect size 0.7) (Table 5.2).

Table 5.2. Mean $_{\pm SEM}$ gaming EI (MJ), time to eating onset during gaming (min) PA METS, energy expenditure (EE) (MJ), gaming relative energy balance (MJ), test meal EI (MJ), total relative energy balance and ingestion time of test meal (min) for all boys (n=21) for each gaming trial.

	Seated video gaming		**Active video gaming**		
	Mean	**SEM**	**Mean**	**SEM**	***p* value**
Gaming EI (MJ)	2.65	0.32	1.63	0.26	<0.001*
Time to eating onset during gaming (min)	7.50	2.32	9.11	2.41	0.811
PA METS	1.22	0.04	1.99	0.11	<0.001*
EE (MJ)	0.39	0.01	0.64	0.03	<0.001*
Gaming relative energy balance (MJ)	2.26	0.32	0.99	0.26	<0.001*
Test meal EI (MJ)	1.08	0.12	1.07	0.10	0.859
Total relative energy balance (MJ)	3.34	0.35	2.06	0.30	<0.001*
Ingestion time of test meal (min)	11.02	4.53	8.48	3.33	0.051

*Indicates a significant difference between the active and seated video gaming trials.

5.3.3.1 Time spent sedentary, in light and moderate to vigorous physical activity

When time spent sedentary, in light and MVPA was examined, it was revealed that the boys spent less time being sedentary during active video gaming (27.38±4.68 min), than seated video gaming (74.45±2.96 min) (p<0.001, large effect size 0.9). Significantly more time was spent in light PA during active video gaming (57.14±4.25 min), than seated video gaming (13.98±2.79 min) (p<0.001, large effect size 0.8). Time spent in MVPA, was significantly greater during active video gaming (5.48±2.61 min), than seated video gaming (0.19±0.88 min) (p=0.008, small effect size) (Table 5.3).

Table 5.3. Mean $\pm_{SEM}$ time (min) spent in sedentary, light PA and MVPA for all boys (n=21), for both gaming bouts.

	Seated no food		**Active no food**	
Time (min) spent	**Mean**	**SEM**	**Mean**	**SEM**
Sedentary	74.45	2.96	27.38[a]	21.44
Light PA	13.98	2.79	57.14[b]	4.25
MVPA	0.19	0.88	5.48[c]	2.61

[a] Significantly, less time was spent being sedentary when active video gaming than seated video gaming (*p*<0.001).
[b] Active video gaming significantly increased the time spent in light PA, in comparison to seated video gaming (*p*<0.001).
[c] Time spent in MVPA was significantly greater during active video gaming, than when seated gaming (*p*=0.008).

5.3.4 Energy intake, relative energy balance, time to eating onset, macronutrient intake and individual foods consumed

Paired t-tests revealed the boys consumed significantly more when seated video gaming (2.65±0.32 MJ), in comparison to when they were active video gaming (1.63±0.26 MJ) (*p*<0.001); small effect, size *d*=0.3) (Figure 5.2 and Table 5.2). Following the seated and active video gaming trials similar amounts of the test meal were consumed (seated video gaming 1.08±0.12 MJ and active video gaming 1.07±0.10 MJ) (*p*=0.913) (Figure 5.2 and Table 5.2).

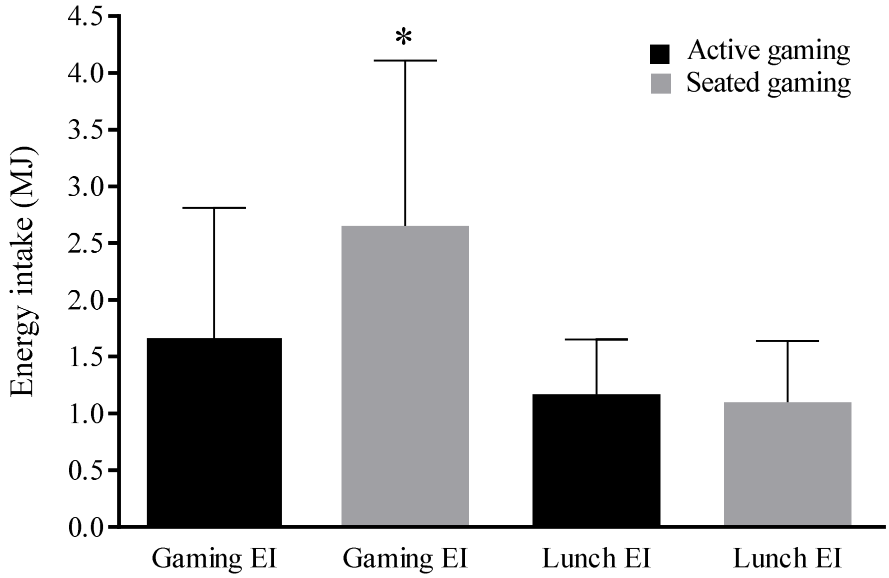

Figure 5.2. Mean $_{\pm SEM}$ gaming and meal EI for all participants (n=21). *Indicates EI during the 90 min seated video gaming significantly greater than during the 90 min active video gaming bout (*p*<0.001).

Following both the active and seated video gaming bouts the boys were in positive relative energy balance. Relative energy balance was significantly greater due to seated video gaming (2.26±0.32 MJ) in comparison to active video gaming (0.99±0.26) (p<0.001, small effect size *d*=0.4) (Table 5.2). When accounting for the test meal EI (MJ), relative energy balance was significantly greater at the end of the seated video gaming trial (3.34±0.35 MJ), in comparison to the active video gaming trial (2.06±0.30 MJ) (p<0.000, moderate effect size *d*=0.6) (Table 5.2).

No significant difference was found in the average time to eating onset (min) (*p*=0.059) between seated and active video gaming (Table 5.2). There was also no difference between the ingestion times of the test meal although there was a trend for significance (*p*=0.051, small effect size *d*=0.3), indicating that the boys took longer to eat the test meal following seated video gaming (Table 5.2).

As a percentage of total EI (MJ), paired t-tests revealed the boys consumed significantly more CHO (58.3±16.7%, *p*=0.004, small effect size *d*=0.3) and protein (6.8±4.4 %, *p*=0.022, small effect size *d*=0.1) but less fat (36.4±14.7 %, *p*=0.004, small effect size *d*=0.3) during the 90 min active video gaming bout, than during 90 min of seated video gaming (CHO 49.3±12; PRO 6.2±2.9 %; fat 44.5±11.3 %) (Figure 5.3).

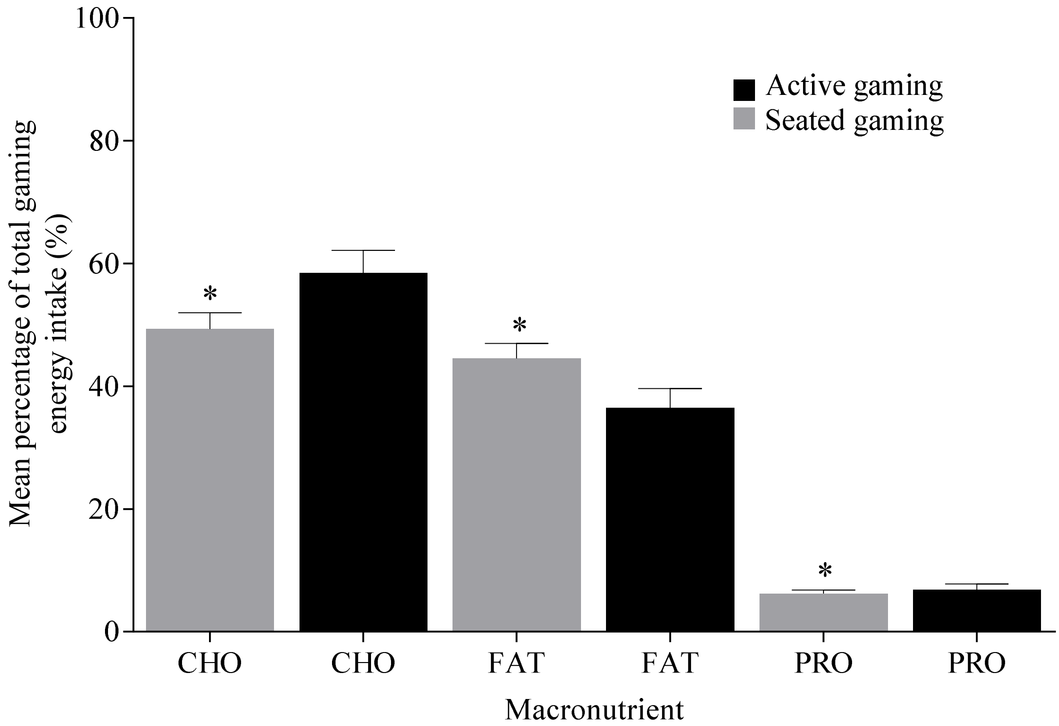

Figure 5.3. Mean amount of macronutrients ingested during gaming for all boys (n=21) as a percentage of total EI. *Indicates the boys consumed significantly more CHO (p=0.004) and PRO (p=0.022) but significantly less fat (p=0.004) during active video gaming than during seated video gaming.

A paired t-test of the individual foods (crisps and apple) and fluids (fruit squash and semi-skimmed milk) consumed during the gaming bouts found most of the energy consumed was from crisps. During active video game play, significantly fewer crisps were consumed (1.10±0.19 MJ) than when seated video gaming (2.04±0.21 MJ) (moderate effect size d=0.5, p<0.05).No significant differences were detected in intakes of apple (active 0.28±0.07 MJ versus seated 0.33±0.10 MJ, p=0.638), fruit squash (active 0.21±0.01 MJ versus seated 0.02±0.01 MJ, p=0.903) or semi-skimmed milk (active 0.23±0.09 MJ versus seated 0.26±0.13 MJ, p=0.992) (Figure 5.4).

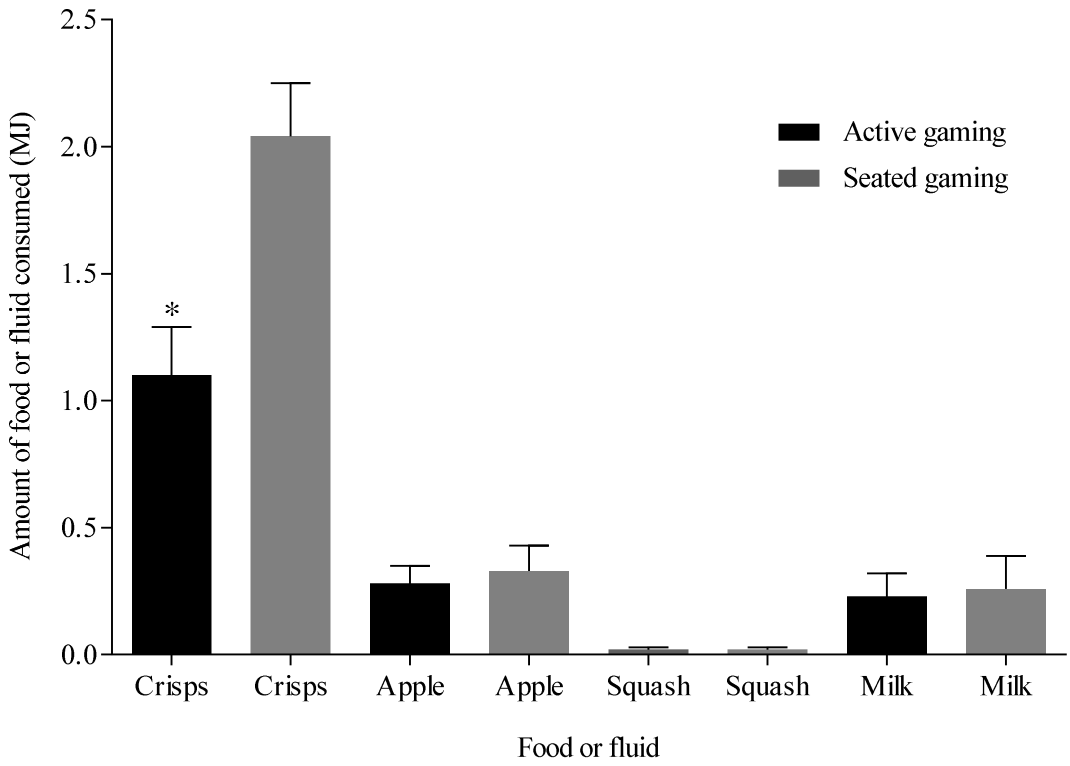

Figure 5.4. Mean amounts of foods and fluids consumed during active and seated video gaming for all boys (n=21). The amount of crisps consumed was significantly lower during active video gaming in comparison to seated video gaming (p<0.000). No significant differences were detected in intakes of apple (p=0.638), squash (p=0.903) or milk (p=0.992) between active and seated video gaming.

5.3.5 Subjective Appetite

Paired t-tests revealed no significant differences in baseline values for sensations of hunger (p=0.917), prospective food consumption (p=0.204) and fullness (p=0.315) between seated and active video gaming. Indicating the boys' sensations of hunger, prospective food consumption and fullness were consistent prior to each gaming trial (Table 5.4).

Table 5.4. Mean $_{\pm SEM}$ subjective appetite sensations of hunger, prospective food consumption and fullness for all participants (n=21) at baseline, time-averaged AUC x 180 min and time-averaged AUC x 90 min for both gaming trials.

		Hunger (not at all hungry = 100 mm)		**Prospective food consumption (nothing at all = 100 mm)**		**Fullness (not full at all = 100 mm)**	
	Gaming bout	**Mean**	**SEM**	**Mean**	**SEM**	**Mean**	**SEM**
Baseline	Seated with food	53	7	46	7	62	6
	Active with food	52	7	51	7	59	8
Time-averaged AUC x 180 min	Seated with food	51	3	52	5	55	4
	Active with food	50	4	48	4	55	4
Time-averaged AUC x 90 min	Seated with food	46	4	46	5	60	6
	Active with food	47	5	45	5	56	5

*Indicates a significant difference between seated and active video gaming.

5.3.5.1 Hunger

Paired t-tests revealed no significant differences in time-averaged AUC x180 min for sensations of hunger (p=0.884) between the active and seated video gaming trials (Table 5.4 and Figure 5.5a). A paired t-test for time-averaged AUC x 90 min for sensations of hunger detected no significant differences due to the active and seated video gaming bouts (p=0.740) (Table 5.4 and Figure 5.5a).

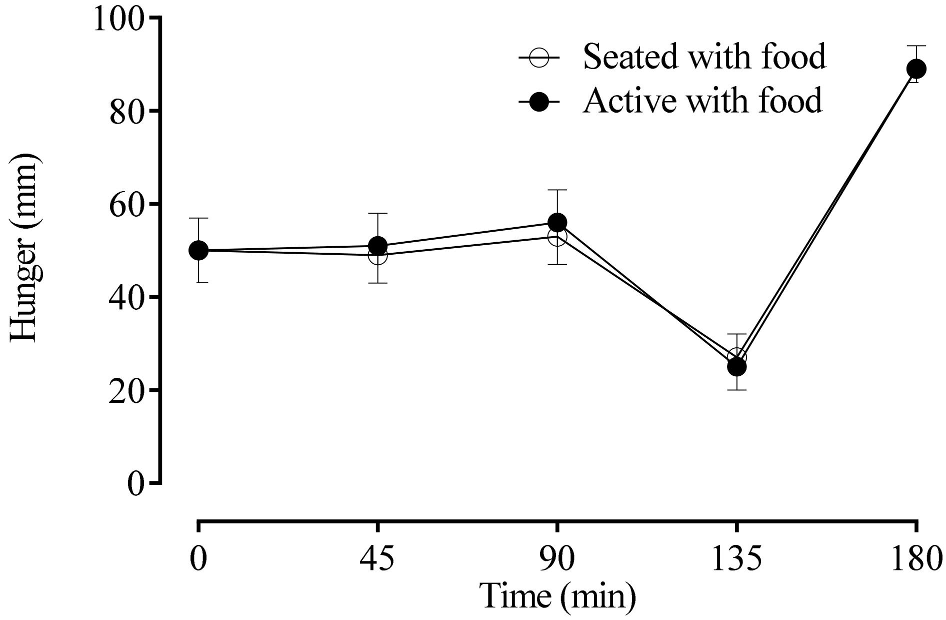

Figure 5.5a. Mean $_{\pm SEM}$ hunger sensations for all participants (n=21) immediately before each active and seated video gaming bout (baseline t=0 min), 45 min during, at the end (90 min) and 45 min post gaming (135 min). No significant difference in time-averaged AUC x 180 min (*p*=0.884) or for time-averaged AUC x 90 min (*p*=0.740).

5.3.5.2 Prospective food consumption

Paired t-tests revealed no significant differences in time-averaged AUC x180 min for sensations of prospective food consumption (p=0.570) between seated and active video gaming (Table 5.4 and Figure 5.5b). A paired t-test for time-averaged AUC x 90 min for sensations of prospective food consumption detected no significant differences due to the active and seated video gaming bouts (p=0.808) (Table 5.4 and Figure 5.5b).

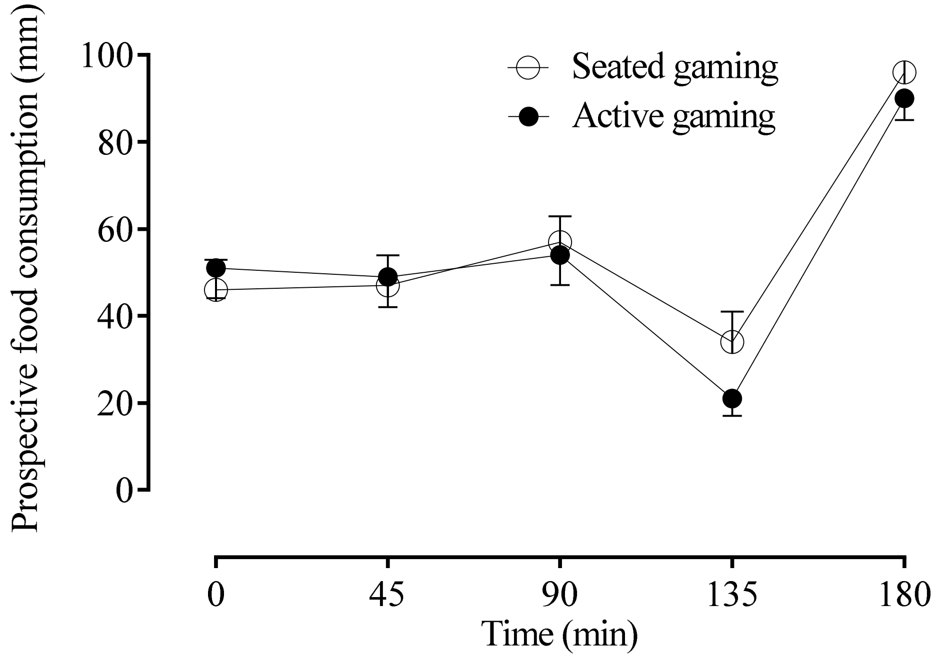

Figure 5.5b. Mean ±SEM sensations for prospective food consumption for all participants (n=21) immediately before each gaming bout (baseline t=0 min), 45 min during, at the end (90 min) and 45 min post gaming (135 min). No significant difference in time-averaged AUC x 180 min (*p*=0.570) or for time-averaged AUC x 90 min (*p*=0.808).

5.3.5.3 Fullness

A paired t-test revealed no significant differences in time-averaged AUC x180 min for sensations of fullness (*p*=0.733) between seated and active video gaming (Table 5.4 and Figure 5.5c). A paired t-test for time-averaged AUC x 90 min for sensations of fullness detected no significant differences due to the active and seated video gaming bouts (*p*=0.379) (Table 5.4 and Figure 5.5c).

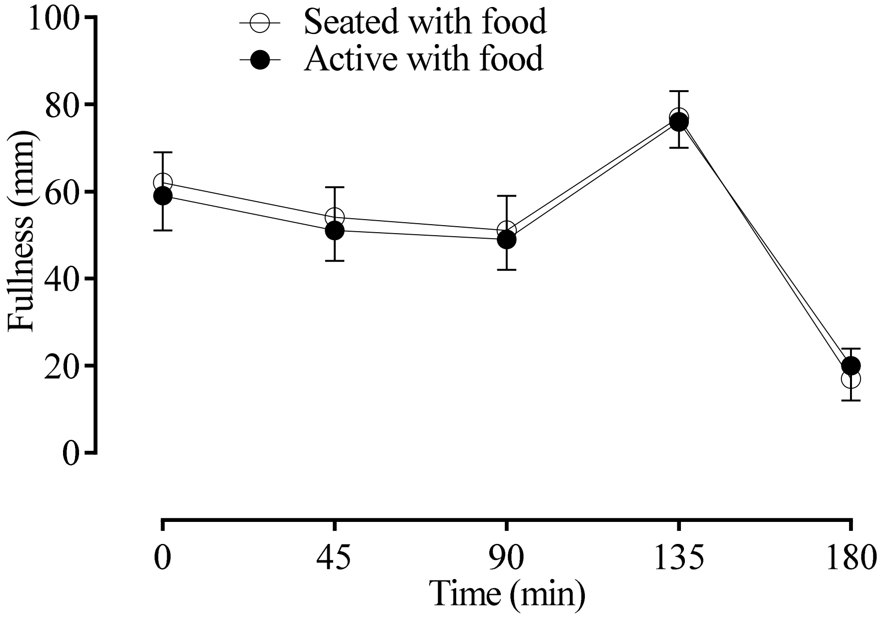

Figure 5.5c. Mean $_{\pm SEM}$ sensations for fullness for all participants (n=21) immediately before each gaming bout (baseline t=0 min), 45 min during, at the end (90 min) and 45 min post gaming (135 min). No significant difference in time-averaged AUC x 180 min (*p*=0.733) or for time-averaged AUC x 90 min (*p*=0.379).

5.3.6 Plasma GLP-1$_{7-36}$ and blood glucose

Paired t-tests detected no significant differences in baseline plasma GLP-1$_{7-36}$ (*p*=0.199) or blood glucose (*p*=0.676) between active and seated video gaming (Figure 5.6a). Paired t-tests revealed no significant differences in time-averaged AUC x 135 min for plasma GLP-1$_{7-36}$ (Figure 5.6a) between active and seated video gaming (*p*=0.413) (Table 5.5). A significant increase was detected in time-averaged AUC x 135 min for glucose during active video gaming (*p*=0.037, small effect, size *d*=0.3) in comparison to seated video gaming (Table 5.5 and Figure 5.6b).

Paired t-tests for time-averaged AUC x 90 min (45 to 135 min) revealed no significant differences for plasma GLP-1$_{7-36}$ between active and seated video gaming (*p*=0.930) (Table 5.5 and Figure 5.6a). Although glucose showed a trend to be significantly greater during active gaming (*p*=0.052) than seated video gaming (Table 5.5 and Figure 5.6b).

Table 5.5. Mean $_{\pm SEM}$ time-averaged AUC x 135 min and AUC x 90 min for plasma GLP-$1_{7\text{-}36}$ (pg/mL) and blood glucose (mmol/L) for each gaming bout for (n=20) boys.

		Active video gaming		**Seated video gaming**		
		Mean	**SEM**	**Mean**	**SEM**	***p* value**
Baseline	GLP-$1_{7\text{-}36}$ (pg/mL)	8.8	1.6	6.3	0.7	0.199
	Glucose (mmol/L)	4.97	0.16	4.91	0.13	0.676
Time averaged AUC x 135 min	GLP-$1_{7\text{-}36}$ (pg/mL)	11.9	1.9	10.4	1.2	0.413
	Glucose (mmol/L)	5.27	0.05	5.03	0.45	0.037*
Time averaged AUC x 90 min	GLP-$1_{7\text{-}36}$ (pg/mL)	12.9	2.1	11.7	1.4	0.930
	Glucose (mmol/L)	5.28	0.07	5.04	0.10	0.052

*Indicates a significant difference between seated and active video gaming.

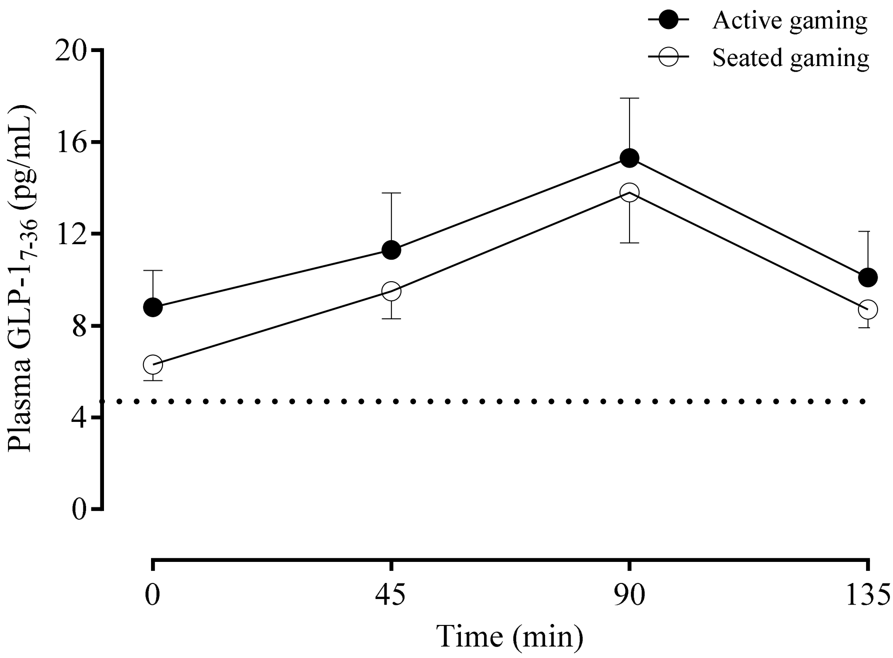

Figure 5.6a. Time-averaged AUC x 135 min (*p*=0.413) and x AUC 90 min (*p*=0.930) for plasma GLP-$1_{7\text{-}36}$ between active and seated video gaming for 20 boys. No significant differences. The dotted line on the graph denotes the fasted levels of GLP-$1_{7\text{-}36}$ of the boys.

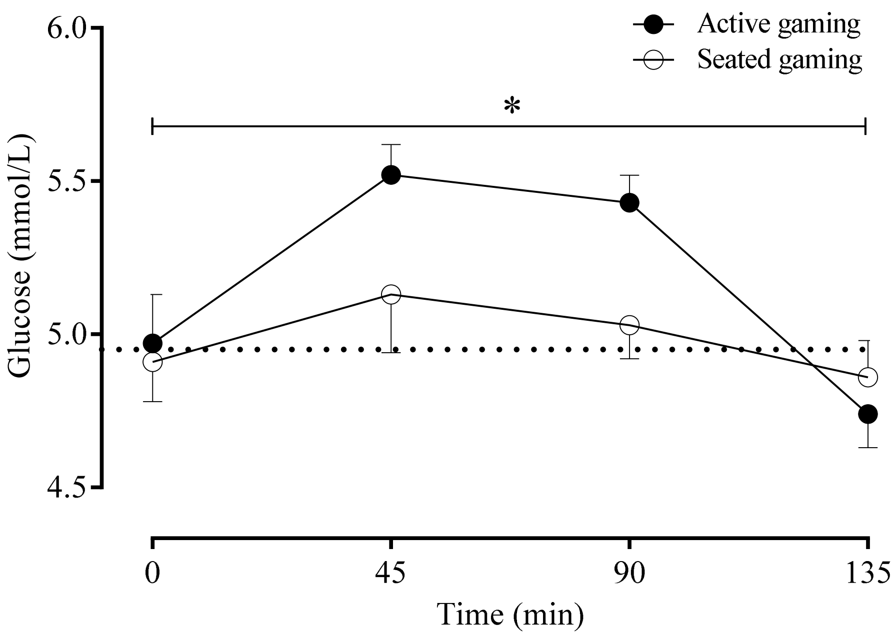

Figure 5.6b. Time-averaged AUC x 135 min and AUC x 90 min blood glucose concentrations, for active and seated video gaming for 20 boys. A significant increase was detected in time-averaged AUC x 135 min blood glucose between seated and active video gaming. Blood glucose was significantly higher during the active video gaming trial than the seated video gaming trial (*p*=0.037). No significant difference in time-averaged AUC x 90 min blood glucose between seated and active video gaming (*p*=0.052). The dotted line denotes the fasted levels of blood glucose of the boys.

5.4 Discussion

The present study was the first to rigorously and objectively measure the acute satiety responses to active and seated video gaming in 8-11 y boys. The homeostatic signals that contribute to the metabolic satiety response, namely plasma GLP-$1_{7\text{-}36}$ and blood glucose were measured simultaneously alongside subjective appetite sensations of hunger, prospective food consumption and fullness and *ad-libitum* gaming EI, during active and seated video gaming. *Ad-libitum* EI from a post-gaming test meal was also measured, to determine whether acute compensation occurred for gaming EI.

The main findings of this study established that time-averaged AUC blood glucose was significantly higher during the active video gaming trial (t=0 min to 135 min), compared to the seated video gaming trial, in 8-11 y boys. However, the *ad-libitum* gaming EI of the boys was significantly greater during seated video gaming, in comparison to active video gaming. Examination of the macronutrients consumed revealed that during the 90 min active video gaming bout there was a greater proportion of CHO consumed, than in the seated video gaming bout. The greater CHO intake was the probable explanation for the higher glucose response observed in the boys when active video gaming. Despite this, no differences were detected in plasma GLP-$1_{7\text{-}36}$, between seated and active video gaming. The largest proportion of energy (MJ) during both 90 min trials was from crisps although, there was a significantly lower amount consumed during active video gaming. Moreover, the greater amounts of food and drink consumed during seated video gaming did not bring about a subsequent reduction in EI in the post-gaming test meal, as intakes were similar following both bouts. Furthermore, when EE was estimated from the sedentary levels of PA elicited by seated video gaming and the light levels produced by active video gaming, this did not offset the EI of the boys during the respective 90 min gaming bouts. At the end of each 90 min gaming bout, the boys were found to be in positive relative energy balance, which was significantly greater from seated video gaming. The relative energy balance became more positive following consumption of the post-gaming test meal in both conditions but was still significantly greater following the seated video gaming trial, than after the active video gaming trial. Subjective sensations of hunger, prospective food consumption and fullness were no different between the two gaming trials. These findings in relation to EI, relative energy balance and subjective appetite sensations are consistent with those found between seated gaming and resting in male adolescents (Chaput et al., 2011) and between seated video gaming and television viewing in 9-13 y boys (Marsh et al., 2013). In addition, 90 min of active video gaming significantly reduced the time the boys being sedentary and increased their time spent in light and MVPA, in comparison to seated video gaming.

The time-averaged AUC concentrations of blood glucose from baseline (t-0 min) to 135 min were greater during the active video gaming trial, than during the seated video gaming trial, even though the boys consumed significantly more when seated. The higher plasma glucose level established at the end of the active video gaming bout (90 min), probably occurred from the greater CHO consumed by the boys relative to total EI. Increased concentrations of blood glucose are in accordance with the 'glucostatic theory' (Mayer, 1952), which is thought to have an essential role in the short-term regulation of appetite. The theory suggests that a rise in blood glucose coincides with an increase in satiety signals which induces satiety and causes suppression of hunger (Mayer, 1952). The higher concentrations of plasma glucose could indicate that satiety was enhanced due to active video gaming. If there was indeed an enhanced level of satiety this could explain the lower EI during the active video gaming bout. Furthermore, although not significantly different, the time-averaged AUC concentrations of plasma GLP-$1_{7\text{-}36}$ for active video gaming were higher than those found during the seated video gaming trial (Figure 5.4a). However the levels of plasma GLP-$1_{7\text{-}36}$ which increased during the 90 min active video gaming bout may not have been strong enough to cause a reduction in EI. This suggests that satiation, the hedonic response to food intake, superseded the homeostatic signal from GLP-$1_{7\text{-}36}$ to stop eating, during active video gaming (Blundell & Finlayson, 2004; Yeomans, 1998).

At the time of writing, no other studies have investigated hormonal signals related to satiety in response to active video gaming in young boys. With regards to seated video gaming there has been only one study with adolescents (Chaput et al., 2011), although it differed in design and appeared to focus on hunger peptides, rather than satiety signals. Direct comparison between the findings of the cited study and those of the present investigation are problematic but nonetheless necessary. The former study of Chaput and colleagues (2011) compared blood glucose, serum insulin, serum cortisol and total plasma ghrelin, during 1 h of seated video gaming versus 1 h of resting. Findings showed that at 40 min during the seated video gaming condition, to cessation of the bout, the plasma glucose concentrations increased significantly in comparison to resting. In contrast, serum insulin although not dissimilar between conditions, was found to decrease significantly over the 1 h period of both seated gaming and resting. However, unlike the present study, food and drinks were not offered *ad-libitum* until 10 min following each of the conditions. Chaput and colleagues (2011) attributed the marked increase in plasma glucose to acute stress caused by the seated video game play, wherein the liver induces the release of the sugar, as in the fight-or-flight response (Dallman, 2010). Despite the proposed acute stress response to seated video gaming, no significant differences were found in serum cortisol between the two conditions and actual levels decreased during the course of both 1 h bouts. Furthermore, the adolescents consumed more in the test meal, post seated video gaming than they did following the resting condition even though total plasma ghrelin levels remained similar. In relation to the increased plasma glucose during seated video gaming, the researchers proposed that according to the 'glucostatic theory', this should have induced satiety but instead it appeared to increase EI. This suggests that the participants eating behaviour was more closely driven by hedonic rather than homeostatic mechanisms (Chaput et al., 2011).

The similarity in findings of Chaput and colleagues (2011) and the present study could indicate that both seated and active video gaming can override food cues that are controlled by assimilated signals from the sensory, neuronal and digestive systems, as also found with television viewing (Temple et al., 2007). As markers of stress were not measured in the present study, the possibility that a stress response had occurred during active video gaming should not be ruled out as this may also account for the higher levels of blood glucose. In addition, the low intensity activity produced by active video gaming would have caused an increased demand for glucose that would have augmented the levels of circulating blood glucose during this bout (Arkinstall et al., 2004). Future research might consider including the measurement of both cortisol and lactate to provide a more accurate depiction of the stress and exercise demand of active video gaming, in comparison to seated video gaming.

In relation to the subjective appetite responses, no differences were detected in sensations of hunger, prospective food consumption or fullness between the seated and active video gaming bouts. Thus far, only two other paediatric studies have investigated the acute effects of active video gaming on subjective appetite (Chaput et al., 2015; Gribbon et al., 2015) and both of these reported similar findings to the present study. The sensations of hunger, prospective food consumption and fullness of healthy male adolescents were no different during 1 h of resting, seated video gaming and active video gaming (Gribbon et al., 2015) or in obese adolescent males during 1 h of resting, seated video gaming, active video gaming and cycling (Chaput et al., 2015). However, the sensations of fullness of the boys during both active and seated video gaming (Figure 5.5c) somewhat reflected the response of plasma GLP-$1_{7\text{-}36}$ (Figure 5.6a). The fullness sensations of the boys increased during the course of the 90 min bouts resulting in them feeling more full at the end of the gaming bouts (90 min) than at baseline (t=0 min), they then felt less full 45 min post-gaming (135 min). The findings of the present study in relation to fullness sensations indicated that satiety may have been instigated during both seated and active video gaming but, the homeostatic signal was not strong enough to override the hedonic response to food intake (Blundell & Finlayson, 2004; Yeomans, 1998).

The total energy consumed by the boys during 90 min of seated video gaming was 2.64±0.32 MJ whilst in the active video gaming bout, EI was significantly lower (1.63±0.26 MJ). Per hour, the values obtained for seated video gaming (1.75 $MJ{\cdot}h^{-1}$) are identical to those found in Chapter IV of this thesis (1.75 $MJ{\cdot}h^{-1}$) although they are higher than those of Mellecker and McManus, (2008), in 8-12 y children (1.57 $MJ{\cdot}h^{-1}$). With regards to active video gaming, in the present study EI was considerably less per hour (1.08 $MJ{\cdot}h^{-1}$) than established in both Chapter IV (1.41 $MJ{\cdot}h^{-1}$) and by (Mellecker & Mcmanus, 2008) (1.60 $MJ{\cdot}h^{-1}$). Nonetheless, the lower EI due to 90 min of active video gaming is still considerable. The differences in EI between active video gaming studies could be due to variations in study design, as the current study was situated in the laboratory as opposed to the after-school setting employed in Chapter IV. In addition and as mentioned in Chapter IV, Mellecker and McManus, (2008) utilised a seated video gaming device attached to a treadmill, instead of a genuine active video gaming console. The mode of active video gaming was therefore, less realistic and as a consequence might have been less stimulating and challenging, which could have caused the children to consume more than observed presently.

Considering the EI of the boys due to 90 min of active video gaming was significantly lower than from seated video gaming, when intakes were compared with the daily EAR for energy, for males aged 9 y, in the UK (7.70 MJ) (Jackson, 2011), substantial amounts of food and drinks had been consumed during both bouts. By the end of the 90 min seated and active video gaming bouts the boys had consumed 34% and 21%, respectively, of the total energy they require per day. Accounting for the estimated energy expended during the bouts, the relative energy balance at cessation of seated and active video gaming was 2.26±0.32 MJ and 0.99±0.26 MJ, respectively. The active video gaming EI therefore was not offset by the extra energy expended during it. Allowing for the post gaming test meal (Table 5.2), at the end of the seated gaming trial the relative energy balance increased to 3.34±0.35 MJ, and 2.06±0.30 MJ at the close of the active video gaming trial. Therefore the boys did not compensate for the extra EI during gaming by down-regulating food intake in the test meal. The resultant relative energy balance at the end of the trials for seated and active video gaming was 43% and 27%, respectively of daily EAR for total EI.

When the individual foods (crisps and apple) and fluids (fruit squash and semi-skimmed milk) consumed by the boys during gaming were examined, it was revealed that 77% of seated and 67.5% of active gaming EI was from crisps. When accounting for the post gaming test meal, crisps were 41% of the total energy consumed during active video gaming and 55% when seated video gaming. In agreement with the findings of Chapter IV, the greatest contributor to the positive, relative energy balance state of the boys which was estimated at cessation of the trials was therefore, crisps. When considering the proposed reduction in daily dietary intake of only 0.46 to 0.69 MJ per day (Wang et al., 2006) the consumption of this particular food during both active and seated game play has the potential to contribute to a positive energy balance state.

The only paediatric study (Gribbon et al., 2015) thus far to have investigated compensation due to active video gaming EE, also did not establish any difference in EI in a post gaming meal, when compared with 1 h of resting and seated video gaming. At the end of the active video gaming trial however, the participants were measured as being in negative energy balance, which was then compensated for 24 h later by an increase in EI (Gribbon et al., 2015). In the cited study however, food was offered *ad-libitum* in a post-trial test meal only and not during the conditions, as in the present study which is the probable explanation for the difference in findings. In the current study, it is not known whether the boys compensated for the extra EI during both gaming trials at a later time, either by a down-regulation in EI or an increase in EE. If no compensation occurred such substantial levels of energy surplus could contribute to a state of positive energy balance which may also be clinically meaningful with regards to weight status. Particularly when a reduction of only 0.46 to 0.69 MJ per day might be all that is required to reduce the energy gap and bring about a decrease in body mass in children (Wang et al., 2006).

In the present study, the average PA METS were significantly greater due to active video gaming (1.99±0.11 MJ) in comparison to seated video gaming (1.22±0.04 MJ). However, the PA METS attained during active video gaming were lower than those found in the previous intervention study in this thesis (Chapter IV) and of former paediatric active video gaming studies that have utilised Nintendo Wii™ Sports tennis (Graves et al., 2008; White et al., 2011). The gaming bouts utilised in the previously published studies were of much shorter duration and so would have enabled a higher level of PA intensity to be maintained. In the present study, the active video gaming bouts utilised were of longer duration and were representative of 7-11 y children from the North East of England. Consequently, the gaming bouts were more true-to life than those used in the previous laboratory studies (Allsop et al., 2013).

The examination of time spent in sedentary, LPA and MVPA revealed there was a shift from sedentary PA in seated video gaming, to light PA in active video gaming which was an increase in PA intensity of 60%. Active video gaming not only decreased the time the boys spent being sedentary but also increased the time spent in both light and MVPA, in comparison to seated video gaming. However, the mean time the boys spent in MVPA was very short (5.48 min) during active video gaming and is similar to the findings of Chapter IV of the present thesis (1.57 min). Nonetheless, active video gaming produced a decrease in time spent being sedentary and increased the time spent in light as the majority of game play was at this latter PA level. The finding reiterates that real-life active video gaming cannot be considered as a complete replacement for other exercise or sports that elicit MVPA. Although, active video game play should be encouraged over seated video gaming to reduce time children spend being sedentary.

Active video gaming in children should be thus be encouraged rather than seated video gaming to help meet PA recommendations (Almond et al., 2011). More specifically, even though 90 min of active video gaming does not attain MVPA, it will contribute to the prevention of sedentary behaviour which is associated with childhood obesity and increased cardio-metabolic health risks (Ekelund et al., 2006; Mitchell, Pate, Beets, & Nader, 2013).

It must be noted that this was the first study to investigate appetite both subjectively and objectively during active video gaming in children and so is not without limitations. Due to the objective measurement of GLP-$1_{7\text{-}36}$, and the very short half-life of the hormone, the study was conducted in the laboratory rather than in a more true-to-life setting, such as school as utilised in Chapter IV. However the change to the gaming environment was necessary, as the laboratory enabled greater assurance in relation to the preservation of GLP-$1_{7\text{-}36}$, than a school-setting would have and essentially, GLP-$1_{7\text{-}36}$, was one of the main variables of interest in the present study. Despite the change to the environment, all other elements relating to the design of gaming bouts remained the same as those established in Chapter III and utilised in Chapter IV. Such continuity is therefore deemed as having been an asset to the study. Furthermore, no measurement of EE during the 1 h rest period prior to the test meal was undertaken however, the boys were resting.

A further strength is that at the time of writing, the present study initiated the investigation of the acute effects of active video gaming on glucose and the satiety-related peptide GLP-$1_{7\text{-}36,}$ in 8-11 y boys. Moreover it was the first study to measure these satiety signals by utilising fingertip capillary blood sampling which was only recently established as being a reproducible and more ethical technique in comparison to antecubital-venous collection (Green et al., 2014). Considering the age of the present study population (8-11 y), this alternative blood sampling technique provided a more suitable method for the collection of blood and thus assessment of GLP-$1_{7\text{-}36}$ and glucose.

To conclude, it was established that an increase in blood glucose occurred during 90 min of active video gaming and one of the likely reasons for this was the greater intakes of CHO in proportion to total EI, in this particular bout. The plasma concentrations of GLP-$1_{7\text{-}36}$ which increased from fasting levels during the active video gaming bout would appear to indicate a satiety response had occurred but this may not have been strong enough to override hedonics and cause a reduction in EI. A hedonic response is evidenced by the positive relative energy balance state at the end of the active video gaming trial which resulted in an energy surplus of 27%. It seems that the hedonic response to food intake superseded the homeostatic signal to stop eating, during active video gaming (Blundell & Finlayson, 2004; Yeomans, 1998). Consequently, if the energy surplus established here was not compensated for later by a down-regulation of EI or an increase in EE, the acute food intake during both active and seated video gaming which was predominantly from the crisps consumed, could contribute to a positive energy balance state. On a more positive note however, the increased time spent in light intensity PA during active video gaming, reduced the time the boys spent being sedentary, in comparison to seated video gaming. Active video gaming might not contribute to MVPA therefore, but could contribute to a reduction in the sedentary time of 8-11 y boys.

The present study findings provide justification for further research which examines the effects of active video gaming on subjective and objective appetite over a longer time period, to establish whether compensation for the extra EI occurs more than 1 h post. Future studies might also consider measuring stress, both subjectively and objectively to determine whether it can be induced by active video gaming and is a contributory factor to the rise in glucose levels. The significantly increased levels of glucose during active video gaming might also have stemmed from the low intensity exercise. As such, further research should include the measurement of lactate to assess the demands of active video gaming (Tolfrey & Armstrong, 1995) and also insulin to determine whether there is suppression of this hormone, which is typical during exercise. It should be noted however that in the preliminary investigation to the present study (Appendix J), insulin was found to have large variation between-days from fingertip capillary blood samples. As such it would be necessary to utilise antecubital-venous sampling for the collection of plasma insulin. Visual analogue scales that measure hedonic food intake should also be utilised. The inclusion of this subjective measurement of hedonic food intake during active and seated video gaming could provide further evidence of a non-homeostatic response, as has been done in the studies of children's television viewing (Temple et al., 2007).

CHAPTER VI

GENERAL DISCUSSION

6.1 Thesis aims

The overarching aim of the present thesis was to investigate the acute EI and appetite responses to real-life active video gaming, in children. To accomplish this aim, the real-life active video gaming practices of 7-11 y children were explored and determined, by questionnaire. Having established the active video gaming practices of children, a study protocol was designed that reflected these. The true-to-life active video gaming protocol was then utilised in both a naturalistic setting and in the laboratory. In the naturalistic setting, the acute appetite sensations and EI of 8-11 y boys were investigated in response to active and seated video gaming. To gain a greater understanding of the EI and appetitive responses of 8-11 y boys to active video gaming, the research in the laboratory, examined the acute metabolic satiety-signals together with subjective sensations and EI. The present chapter will therefore, discuss and appraise the findings of the experimental research conducted and described in Chapters III to V of this thesis, reflect on the study methodology utilised and provide directions for future active video gaming research and practice.

6.2 Reflections on main findings

6.2.1 The real-life active video gaming practices of children

The studies described in Chapters III to V of this thesis evolved due to the introduction of active video games to the video gaming arena (Lofgren, 2015) and the potential detrimental effects of sedentary screen based media to children's PA, sedentary behaviour, EI and weight status (discussed in section 2.4). Chapter III in particular, was developed from research which explored whether active video gaming could be used to increase children's PA. Findings from these previous studies revealed that when compared with resting, sedentary screen-based media, seated video gaming and walking on a treadmill, children could expend two to three times more energy and elicit two to six METS through active video game play (Lanningham-Foster et al., 2006; Maddison et al., 2007; White et al., 2011). The majority of these earlier studies however, did not provide justification for aspects relating to study design. These included the time-period of gaming bouts, the gaming console(s) and game(s) utilised, whether the games were played against other individuals or indeed, as previously evidenced with sedentary screen based media activity (Berentzen et al., 2014; Borghese et al., 2014; Pearson & Biddle, 2011), whether food and (or) drinks were consumed during play. These aforementioned aspects of active video game play could have important implications on the levels of PA attained and energy balance in children. Evidence then began to emerge from studies which surveyed the active video gaming behaviours of adolescents, particularly in relation to the gaming console and active video games they played and the typical duration of game play (O'Loughlin et al., 2012; Simons, Bernaards, et al., 2012; Song et al., 2015). It became clearer that there was a discrepancy between the majority of the previous laboratory-based studies and survey findings, as the timing of the bouts utilised were of a shorter duration, than reported by adolescents. Thus emphasising the importance of investigating active video gaming practices first, to help design an appropriate protocol that could be utilised to examine acute appetite and EI, in response to gaming protocols.

At the time the present research was implemented, no surveys of children's active gaming practices had been conducted in the UK. On this premise and due to the gaps in knowledge in active video gaming described above, it was deemed essential in Chapter III to develop a questionnaire (Appendix A) that enabled the active video gaming practices of 7-11 y (key stage two) to be established. Following the development and piloting of the 'Active Gaming Questionnaire' Appendix A), a survey of the active video gaming practices of a subsample of 7-11 y children from Newcastle upon Tyne, was implemented. The resultant main findings of 'The real-life active video gaming practices of 7 to11 y children revealed, that they had good accessibility to active video games. They were viewed by the children and their parents (main carer) as being an alternative form of exercise which was beneficial due to the '*physical effects'* they observed during game play and that were perceived as having '*health benefit*s'. The games were played for these '*health* *benefits*', for the '*entertainment'* value and often due to the children living in an '*unsafe environment'* that restricted outdoor play (Allsop et al., 2013).

The most popular gaming console and active video game played by the children was established as the Nintendo Wii™ and the Nintendo Wii™ Sports compendium. The children also liked to play active video games against other individuals. The majority of children consumed food and (or) drinks during active video game play and when they did, they typically played for an average of 81±50 min, which was longer than reported by the non-consumers of food and (or) drinks) (57±32 min). The most popular foods consumed during active video gaming were fruit and crisps and the most popular drinks were fruit flavoured juice and milk. Furthermore, most of the respondents were boys (72.5%) aged 9±1.3 y (Allsop et al., 2013). These latter findings enabled a gaming protocol to be designed that could facilitate the investigation of the acute EI and appetite responses to real-life active video gaming, in 8-11 y boys and which was utilised in Chapter IV and Chapter V.

6.2.2 Acute energy intake responses to active gaming

In view of the findings of Chapter III that the majority of children consumed foods and/or fluids whilst they played active video games and the preliminary evidence of EI during sedentary screen based media use (Chapter II, section 2.4.1), the exploration of EI and relative energy balance became key to the thesis. In Chapter IV, the acute subjective appetite and EI of 8 to 11 y boys during active and seated video gaming was investigated by utilising four individual gaming bouts. The four gaming bouts were: 1) 90 min seated video gaming, no food or drinks offered; 2) 90 min active video gaming, no food or drinks offered; 3) 90 min seated video gaming with food and drinks offered *ad-libitum*; 4) 90 min active video gaming with food and drinks offered *ad-libitum*. In the two gaming bouts in which food and drinks were offered *ad-libitum*, no significant differences were found in EI between active (2.14±0.28 MJ) and seated video gaming (2.84±0.26 MJ). When calculated as a proportion of daily EAR for energy, for boys aged 9 y in the UK (7.70 MJ per day) (Jackson, 2011), the EI during active and seated video gaming was 27% and 34%, respectively. The EI during the active video gaming bout was not counterbalanced by the energy expended during it. Consequently, at the end of the 90 min gaming bouts, the relative energy balance of the boys was 1.50±0.27 MJ for active video gaming and 2.37±0.33 MJ for seated video gaming. If not compensated for later, the EI observed during both active and seated video gaming could therefore contribute to a chronic positive energy state.

As it was not known whether the boys compensated for the EI during the bouts at a later time-point, this provided justification for Chapter V to extend the gaming trials to include a post-gaming test meal. The addition of the test meal however, resulted in an increase in the relative energy balance state of the boys. During the 90 min bouts, the boys were once again observed to consume a significantly greater amount when seated (2.65±0.32 MJ) than when active video gaming (1.63±0.26 MJ). One hour later, in the test meal however, they consumed similar amounts of food (active video gaming 1.07±0.10 MJ, seated video gaming 1.08±0.12 MJ). When allowing for the gaming EE therefore, as in Chapter IV, they did not compensate for the EI during active or seated video gaming. Consequently, relative energy balance increased from 2.26±0.32 MJ to 3.34±0.35 MJ in the seated trial and from 0.99±0.26 MJ to 2.06±0.30 MJ in the active video gaming trial. When the individual foods consumed during the 90 min gaming bouts were examined, it was revealed that most of the EI was from crisps. However, it is noteworthy that the amount of foods and fluids consumed was decreased by 10% in response to active video gaming this also resulted in significantly lower relative energy balance when compared to seated video gaming.

The EI and relative energy balance findings are particularly important in view of the proposal that a reduction in EI by 0.46 to 0.69 MJ per day, could be all that is required for overweight or obese children to achieve energy balance (Wang et al., 2006). The relative energy balance of the boys determined in both Chapters IV and V exceeded the proposed reduction of Wang and colleagues (2006) and could therefore, be clinically meaningful in relation to body mass. Children who play both active and seated video games on a daily basis and eat and (or) drink during game play, especially snacks such as crisps which was the food from which most energy was found to be derived, could be more vulnerable to a positive energy state. Those who play active video games due to the surrounding environment being unsuitable for outdoor play, (as established in Chapter III), might play them more, and so may be at an even greater risk.

6.2.3 Acute subjective appetite responses

As described in Chapter II, section 2.5, appetite and the subjective components, namely hunger, prospective food consumption and fullness are influential upon EI. At the time the investigation described in Chapter IV was implemented, no other studies had examined paediatric subjective appetite responses to active video gaming. Chapters IV and V demonstrated that the acute subjective appetite sensations of hunger, prospective food consumption and fullness, during seated and active video gaming were no different when food and drinks were offered *ad-libitum*. The only difference in subjective appetite sensations was detected in Chapter IV, when food and drinks were not offered during one active and one seated video gaming bout. In comparison to the two bouts that offered food and drinks *ad-libitum,* the boys reported feeling more hungry, wanted to eat more and were less full during both active and seated video gaming. This would seem an unsurprising finding since the boys were asked to refrain from eating and drinking following their standardised lunch (1230 h) until cessation of the bouts (1700 h). Considering, feelings of hunger and prospective food consumption were reduced and fullness sensations were raised during the bouts with food and fluids, the boys still consumed an average of ~ 30% of the daily EAR. The investigation of acute subjective appetite sensations therefore did not provide explanation for the substantial EI during active and seated video game play.

In accordance with the findings of Chapters IV and V, two recent studies also detected no differences in sensations of hunger, prospective food consumption and fullness between active and seated video gaming, or indeed in comparison to any of the other conditions utilised, which included resting and cycling, in the adolescent groups studied (12-15 y and 13-17 y males) (Chaput et al., 2015; Gribbon et al., 2015). Furthermore, the two cited studies observed no differences in acute EI following active and seated video gaming, although it must be noted that unlike in Chapters IV and V, food was not offered during, but after the bouts.

The subjective appetite findings of Chapters IV and V are similar also to those of seated video gaming studies which found no differences in sensations of hunger, prospective food consumption and fullness, in comparison to resting and watching television (Chaput et al., 2011; Marsh et al., 2013). The two seated video gaming studies also observed considerable levels of EI during and following the bouts (Chaput et al., 2011; Marsh et al., 2013). In view of the similarities in findings, it would seem reasonable to suggest that as with television viewing, both seated and active video gaming might have disrupted sensory, neuronal and digestive signals (Temple et al., 2007). Consequently, the drive to eat was sustained (Finlayson et al., 2008; Yeomans, 1998) thus, leading to over-compensation in EI for EE in both gaming bouts. As this hypothesis could not be ascertained by subjective investigation, further and more objective study was therefore necessary, to establish whether appetite signals were indeed disrupted.

6.2.4 Satiety-related signals

The study described in Chapter V, was developed to investigate appetite objectively so as to try and determine whether homeostatic appetite signals in 8-11 y boys are disrupted during active and seated video gaming. To the best of my knowledge, the study was the first in paediatric active video gaming to include the measurement of satiety-related signals. To understand some of the more commonly studied satiety-related signals, specifically GLP-$1_{7\text{-}36}$, glucagon, leptin, insulin and glucose more clearly, the between-day reproducibility of these hormones was established first. In doing this, if an increase or decrease in concentration from fasting was detected in any of the signals during active or seated video gaming, it could be confirmed that this was due to the activity and not typical error. The novel method of fingertip blood sampling was utilised, which had only recently been assessed as being as reproducible as the more invasive antecubital-venous sampling (Green et al., 2014). Following the collection of two fasting samples of capillary blood from the fingertips of 21, 8-11y boys, reproducibility testing showed good, between-day agreement for GLP-$1_{7\text{-}36}$ and glucose. The initial reproducibility testing thus enabled the objective measurement of the satiety-related signals GLP-$1_{7\text{-}36}$ and glucose, to be implemented with accuracy.

The objective measurement of GLP-$1_{7\text{-}36}$ and glucose alongside the subjective assessment of hunger, prospective food consumption and fullness and acute EI, enabled a more rigorous investigation of appetite during active and seated video gaming. The objective study of appetite outlined in Chapter V found no significant differences in plasma GLP-$1_{7\text{-}36}$ responses between active and seated video gaming in the 8-11 y boys. Yet there was a difference detected for glucose which increased significantly during active video gaming. Inspection of macronutrient intakes revealed that even though EI was greater during the 90 min seated video gaming bout, the amount of CHO in proportion to the total energy consumed was significantly greater during active video gaming.

There are nonetheless other possible explanations for the higher glucose levels during active video gaming. According to the 'glucostatic theory' (Mayer, 1952), an increase in blood glucose is indicative of a satiety response. In support of this, the plasma concentration of GLP-$1_{7\text{-}36}$, although not significantly different between the two bouts, was higher during active video gaming and had risen above pre-determined fasting levels (Appendix J). When considering the boys consumed 21% of total daily EAR during active video gaming, it would appear that the homeostatic signals were not able to override the hedonic food cues. The resultant acute effect at the end of the active video gaming intervention was the boys were in positive relative energy balance.

Active video game play may also have incurred a greater amount of stress which is symptomatic of the 'fight or flight' response and causes the release of glucose into the bloodstream (Temple et al., 2007). Furthermore, the low intensity exercise elicited by active video gaming would have caused an increased demand for glucose in the muscles (Arkinstall et al., 2004). The findings in relation to the objective measurement of the satiety-related signals, of GLP-$1_{7\text{-}36}$ and glucose were therefore, not definitive. For this reason, further objective investigation of satiety-related signals is required that incorporates the measurement of hormones which reflect stress and muscle metabolism during exercise.

6.2.5 Active video gaming physical activity

In chapters IV and V, PA during active and seated video gaming was measured by accelerometry. The measurement of PA by accelerometry during active and seated video gaming was a further essential component of the present thesis, for two reasons. Firstly, it enabled the PA levels (METS) achieved by the boys during active and seated gaming to be evaluated with existing PA recommendations. Secondly, the measurements (METS) could be utilised to estimate the EE of the boys during the gaming bouts and thus their relative energy balance.

The measurement of PA established that for the majority of time during the 90 min seated video gaming bout, the boys were sedentary. A more encouraging finding was that, during active video gaming, the boys spent the majority of time in light PA. These PA findings were similar to those of previous studies that had also utilised the Nintendo Wii™ and Nintendo Wii™, Sports tennis (Graves et al., 2008; White et al., 2011). In addition, in Chapter V a significant increase was observed in the amount of time spent in MVPA during active video gaming (active video gaming 5.48 min; seated video gaming 0.19 min) although, this would not make a major impact on time that is recommended to be spent at this PA intensity.

Although the change in PA intensity found presently was only from sedentary during seated video gaming, to light during active video gaming, it was equivalent to an average increase by ~52% PA METS. Furthermore, for ~70% of the time, the boys were being physically active at light intensity levels during the 90 min of active video gaming whilst when seated video gaming, they were sedentary. In view of PA guidelines, this is an important increase, as even though active video gaming with Nintendo Wii™ and Nintendo Wii™, Sports tennis would not help achieve MVPA, the results of Chapters VI and V indicated that it could be used to reduce sedentary activity (Almond et al., 2011). Children should therefore be encouraged to play active video games rather than seated video games and refrain from other sedentary screen-based activities as a form of recreation.

6.3 Methodological limitations

Specific limitations relating to each study have been outlined in the individual experimental chapters (Chapters III, IV and V) of this thesis. The purpose of the present section therefore, is to discuss general limitations in relation to the chosen study population and the methodology utilised in this thesis.

6.3.1 Conducting research with children

Conducting a research project with primary school-aged children presented certain challenges which were firstly encountered with recruitment and then data collection. The best way to gain access to the children in order to carry out the research was believed to be through primary schools. In total, 17 primary schools in and around the city centre of Newcastle upon Tyne were approached, initially with a letter which was followed up with both telephone calls and emails. Out of the 17 primary schools identified, there were only three Head Teachers in Chapter III, two in Chapter IV and one in Chapter V that showed interest in the research project and agreed to a meeting. During the meeting, a detailed outline of the studies was provided. In order to obtain the consent of the Head Teachers, it was necessary to negotiate access to the children so that the studies would not be a major interference to the normal school day and this invariably meant that study protocols had to be slightly adapted. Once consent was granted by the Head Teachers, access was given to meet with the pupils and their parents wherein a further challenge was to obtain consent from them to participate. Both parents and children typically have work, family and social obligations. Participation in research may have been considered as an extra burden on busy lives and is a probable explanation for why many of the children and parents who were approached, unfortunately declined the invitation to take part.

As well as recruitment there were difficulties encountered with data collection, particularly in the studies described in Chapters IV and V. In these studies, it was necessary for the 8-11 y boys under the supervision of their parent or main carer, to replicate their food and fluid intake from 1700 h the previous day, until their arrival at school on the day of the gaming intervention. The boys were also asked to complete a food diary as evidence of food and fluid replication. In addition to this, in Chapter IV, at 15 and 30 min following each gaming bout, the boys were asked to rate and record their subjective appetite sensations in an appetite diary. To ensure that food and fluid intake was replicated and the food and appetite diaries were completed, the children and parents were given reminders either by email, text message or telephone. There were however, some instances where food and fluid intake was not replicated and food and appetite diaries were not completed. It was then necessary to re-test some of the boys and this resulted in the data collection process taking longer than was originally anticipated. In addition, food and fluid intake was not replicated by two of the boys and so one of them had to be omitted from the analysis (Chapter V) and another had to be excluded half-way through data collection, due to disruptive behaviour (Chapter IV).

As a consequence of the challenges encountered with recruitment and data collection, the sample size for the studies conducted and described in Chapters III, IV and V, was limited to children from the city of Newcastle upon Tyne. Furthermore, the sample sizes in all of the studies conducted in this thesis were small however, those in Chapters IV and V are similar to previous paediatric appetitive active video gaming investigations (Chaput et al., 2015; Chaput et al., 2011; Gribbon et al., 2015). The research findings and particularly those in Chapter III therefore, are specific only to the children who participated in the studies and are not generalizable to other populations. Consequently, future investigations of the real-life active gaming practices of children would benefit from a greater number of participants. The real-life active gaming survey should be extended to the wider population of UK children as this would be more representative of 7-11 year olds (Bartlett, Kotrlik, & Higgins, 2001). However, it should be noted that the city of Newcastle upon Tyne has a socio-demographic structure which typically reflects the UK population (White et al., 2004). It is often referred to as a microcosm of the UK and as a consequence the city is often chosen as a preliminary location for research (White et al., 2004).

6.3.2 Study Designs

In the present thesis, various study designs were employed and a systematic approach was utilised. First of all the research required the development and use of an Active Gaming Questionnaire (Appendix A). The questionnaire was implemented in a cross-sectional survey, to establish the real life active gaming practices of 7-11 y children (Chapter III). The questionnaire incorporated questions that mostly required quantitative analysis, although some were examined qualitatively. Once the real-life active gaming practices had been established, randomised crossover trials that were as naturalistic as possible, were then implemented to explore the EI, appetite and PA levels of 8-11 y boys, in response to active video gaming. A strength of the present thesis is the systematic approach that was used to carry out the research however, there are additional aspects of the studies which require further discussion.

In Chapter IV and Chapter V, the gaming interventions were conducted in primary schools and the University laboratory, respectively. In doing this, the true-to-life aspects such as playing active video games at home or at a friend's home, were somewhat removed. However, to obtain measures of EI, appetite and PA which were controlled for, it was essential to be able to observe the boys during the gaming bouts.

During the 90 min active video gaming bouts the boys were permitted to play one specific active video gaming console (Nintendo Wii™) and active video game (Nintendo Wii™ Sports, tennis) only. It is acknowledged that 90 min seems to be a long time to play one specific active video game and this may have led to the boys being less engaged than when playing the seated video game. If there was a reduced level of engagement this may have had a lowering effect on PA and EI. However, it enabled more accurate comparison between the active video gaming bouts in Chapter IV.

As only one active video game was permitted to be played by the boys, the findings presented in Chapters IV and V cannot be applied to all active video gaming consoles and games. Indeed, since April 2012 when the first study was implemented (Chapter III) and established Nintendo Wii™ Sports, tennis to be the favourite active video game in the study population, the sales of Xbox 360 Kinect, as of June 2014 had reached 84 million. With the introduction of Xbox One in November 2013 which can also integrate the Kinect sensor, these consoles and games may now be more popular and accessible to 8-11 y boys. One notable advantage of Xbox 360 Kinect is that it does not require a handheld controller and so permits a greater range of movement and so greater EE will likely be elicited. A drawback of the Nintendo Wii™, which was identified as the most popular active gaming console in Chapter III is, that it requires the use of a handheld controller, which can induce the player to flick the wrist, rather than move the whole body. However throughout the gaming bouts, when playing Nintendo Wii™ Sports tennis, the boys were requested not to flick the controller.

A further limitation was the short time scale of the studies in Chapters IV and VI. With the help of a parent, the boys were required to abstain from PA record and replicate all food and fluids consumed from 1700 h the day prior to each intervention, until arrival at school the next morning. Following each gaming intervention, the boys were no longer requested to abstain from PA or record and replicate food and fluid intake. A drawback of this however, was that the measurement of overall energy balance over a 24 h period was precluded and so if there had been a compensatory effect of the EI during the gaming bouts later that day, it was not observed. Even though in Chapter V, the boys did not compensate for the gaming EI at the post-gaming test meal, it cannot be assumed that an up-regulation of EE or down-regulation of EI, may have occurred later. The findings which allude to energy balance should therefore be interpreted with caution. However, to ask the boys and their parents to record food and fluid intake would have placed extra burden upon them. Extra burden had the potential to increase the number of boys who withdrew from the studies and who failed to replicate food and fluid intake. As such, further recruitment and testing, as well as exclusion from statistical analysis might have been necessary.

6.3.3 Accelerometry

In the planning and data collections stages of this research project, a great deal of consideration was given to whether accelerometry was the most appropriate method of EE measurement. It was acknowledged that the estimation of EE by accelerometry is not as precise as indirect calorimetry or DLW, the latter being considered the 'gold standard'. The measurement of PA by accelerometry, subsequent data analysis and conversion to EE results in lower accuracy, typically observed as underestimation (Rowlands, 2007). However, by utilising accelerometry, PA could be classified into sedentary, light, moderate, moderate to vigorous and vigorous levels and the time (min) spent at these various levels could be calculated. A major advantage of classifying PA and calculating the minutes spent at each level from the accelerometry data was that there could be direct comparison to UK Government recommendations for children's PA (Bull et al., 2010). Both DLW and indirect calorimetry do not allow such comparison.

A further consideration with accelerometry was the length of epoch used while recording the PA date. In the present study, an epoch of 10 s was utilised and so it is possible that if there was any vigorous PA during gaming, this might have been misclassified as lower levels of activity and so a shorter epoch may have provided greater accuracy. Indeed, when utilising accelerometers in paediatric populations, it is generally believed that shorter epochs such as 1 s are more accurate, given that children's PA is highly sporadic (Reilly et al., 2008). However, when 1 s epochs were previously compared with 60 s epochs in 7-11 y children, only very vigorous PA was found to be affected and there was no significant effect on light or moderate PA (Reilly et al., 2008). As for the majority of time the PA of boys was classified as sedentary when seated and light during active video gaming, the 10 s epoch used presently may not have significantly reduced the accuracy of the active video gaming EE estimations.

A further consideration was the placement of the accelerometer device and whether this should be on the hip or wrist. Given that active video gaming is predominantly upper limb rather than whole body movement, it was envisaged that wrist placement would provide the most accurate measurement of PA and subsequent estimation of EE. Throughout data collection in Chapter IV therefore, an accelerometer was placed on the right hip and each wrist of participants. Analysis of the data revealed that accelerometer counts from the wrists were extremely high and resulted in PA METS for active video gaming being classified as very vigorous. This meant that PA METS for active video gaming were higher than for sports such as football and basketball (Graves et al., 2008; Ridley & Olds, 2008) and researcher observation of the participant game play, judged this to be improbable. For that reason, wrist data was excluded from the data analysis in Chapter IV and wrist accelerometry was not utilised at all in Chapter V.

Although the measurement of PA and subsequent estimation of EE by accelerometry has several limitations, the method enabled the real-life aspects of children's active video gaming to be retained. Specifically, the participants were able to play Nintendo Wii™ Sports, tennis together and move about freely, with minimal burden and stress which might have been induced with indirect calorimetry.

6.3.4 Energy intake

A limitation in relation to EI, during the active and seated video gaming bouts in Chapters IV and V, was the restricted variability of foods provided to the boys. If there had been more variety, the boys may have taken a greater amount of time to choose the food or fluid they wished to consume, which might have reduced the time they spent in game play. It was felt that it would be more pragmatic to provide only two foods and two fluids. Consequently, apple, crisps, fruit squash and semi-skimmed milk were selected, as they were the most popular foods and fluids consumed by the 7-11 y children, who reported eating and (or) drinking during real-life active video game play (Chapter III). Although it is acknowledged that the foods and fluids, specifically the apple and semi-skimmed may have been reported for reasons associated with social desirability. Nonetheless, the two specific foods and fluids offered *ad-libitum* were likely to be snacks available in the home and as such contributed to the naturalistic approach that was intentionally adopted in the present thesis.

During the gaming bouts employed in Chapters IV and V, the boys were randomised in pairs to play either the active video game (Nintendo Wii™) or the seated video game (Nintendo 3DS). The foods and fluids offered *ad-libitum* during the gaming bouts were placed at a station designated to each boy. By offering the food and fluid items in this way, the boys may have influenced one another to take food or fluid during the breaks that naturally and intermittently occurred between points and games. It has been found in children that EI can be affected due to low acceptance by peers and this can lead to a reduction in consumption of healthy snacks such as fruit (Finnerty, Reeves, Dabinett, Jeanes, & Vögele, 2010). In an effort to minimise any potential peer influence, the pair who were assigned to the Nintendo Wii™ played together against the computer. The two boys who played the Nintendo 3DS played individually, against the computer. In doing this, any peer influence which may have occurred due to winning or losing was reduced, along with subsequent effects on EI and appetite responses.

Differences in EI during active video gaming were observed between the primary school (Chapter IV) and laboratory (Chapter V) based settings. It was noted by the researcher that the boys appeared to be more relaxed in the primary school setting, which was most likely due to it being a more familiar environment to them. In the laboratory, the space was arranged to resemble a kitchen in the home, as far as possible and the blood sampling area was hidden from view during gaming. Irrespective of these attempts to make the laboratory setting more relaxed, there may still have been an element of anticipation in relation to the blood sampling which might have led to the reduction in active gaming EI and PA observed in Chapter V, in comparison to Chapter IV. The amount of foods and fluids the boys consumed when they were active gaming in the primary school (2.30 MJ) in Chapter IV, was greater than when the intervention was situated in the laboratory (0.99 MJ). In addition, the PA levels were greater in the primary school than in the laboratory (2.08 METS and 1.99 METS, respectively). Despite the higher PA levels attained in the primary school setting, the relative energy balance of the boys (primary school 1.64 MJ) was 0.65 MJ greater than in the laboratory (0.99 MJ). As a result, the more relaxed environment of the primary school appeared to have an effect that could be clinically meaningful in terms of body mass (Wang et al., 2006).

6.3.5 Subjective appetite responses

In Chapters IV and V, the study samples were made up with boys of lean, overweight and obese body mass. It is acknowledged that body mass may affect the subjective appetite sensations of children, in response to imposed exercise. However, the low numbers of overweight and obese boys prevented statistical analysis according to body mass as this would have likely produced skewed findings. A recent review of subjective appetite responses to EE in lean, overweight and obese paediatric groups revealed that findings appeared to vary greatly (Thivel & Chaput, 2014). In lean children, there have been increases, decreases and no changes observed in hunger and prospective food consumption with the majority of studies showing no alterations in fullness or subsequent EI (Bellissimo et al., 2007; Bozinowski et al., 2009; Dodd et al., 2008; Thivel & Chaput, 2014). Although in girls, both an increase (Rumbold et al., 2011) and a decrease in EI (Moore, Dodd, Welsman, & Armstrong, 2004) have been detected.

Investigations into appetite responses to EE, with overweight and obese children have largely shown an increase or no change in both hunger and prospective food consumption and for fullness, either a decrease or no change (Dodd et al., 2008; Tamam et al., 2012; Thivel & Chaput, 2014). Similar to lean children, subsequent EI was largely unchanged, although decreases were observed in obese males (Thivel & Chaput, 2014; Thivel, Metz, Julien, Morio & Duché, 2014). It was suggested by Thivel and Chaput (2014), that the variation in subjective appetite responses may be due to the differing types, duration and intensities of exercise utilised in the aforementioned studies (Thivel & Chaput, 2014). However, the age of the paediatric populations of the investigations included in the review varied considerably from children aged 8 y (Nemet et al., 2010), up to adolescents aged 15 y (Thivel et al., 2012). The 8 y old children would have been at a far earlier stage of maturation than the 15 y adolescents, which may be a further reason for the variation in findings between studies (Thivel et al., 2011). There is a small amount of evidence that suggests the plasma concentrations of some appetite-related hormones differ according to maturation status (Horner & Lee, 2015). Physiological appetite responses according to maturation could therefore lead to variations in appetite sensations in differing paediatric populations (Horner & Lee, 2015; Thivel & Chaput, 2014).

In relation to sex, the present thesis, limited the study of appetite responses to active video gaming, to boys. To date, no active video gaming studies have compared the appetite responses of boys and girls. With regards to paediatric appetite studies, very few have examined the responses from imposed exercise and thus far, findings are equivocal. In response to short duration exercise, an increase in hunger and PFC and a decrease in fullness were observed in both lean boys and girls (Bozinovski et al., 2009). However, the increase in hunger and subsequent EI at a test meal was significantly lower in the boys, than the girls (Bozinovski et al., 2009). When the responses of obese boys and girls were examined following a bout of acute cycling, no sex differences were observed in subjective appetite or subsequent EI (Thivel et al., 2011). In comparison to a sedentary condition, EI was decreased by exercise in both sexes (Thivel et al., 2011). More recently, the appetite responses of lean and obese boys and girls were compared following a bout of acute cycling (Thivel et al., 2014). In relation to body mass, EE and EI were significantly increased in the obese boys and girls in comparison to the lean children, yet no differences were observed in hunger, PFC or fullness. With regards to sex, there were no differences in appetite or subsequent EI between the obese and lean boys and girls (Thivel et al., 2014). Whether these differences in appetite findings are due to body mass or sex, is as yet unclear (Thivel & Chaput, 2014).

To date only one study has examined appetite and EI in response active video gaming according to body mass (Chaput et al., 2015). As with seated video gaming, moderate intensity cycling and a sedentary condition, hunger and PFC increased, fullness decreased and EI was significantly higher in obese adolescent males, than in those with lean body mass (Chaput et al., 2015). These recent findings and those of the aforementioned paediatric studies demonstrate that further research which compares appetite responses according to children's body mass and sex, in response to active video gaming are warranted.

6.3.6 Measurement of satiety-related signals

In Chapter V, there was a preliminary study to assess the between-day reproducibility of fasting GLP-$1_{7\text{-}36}$, glucagon, leptin, insulin and glucose by fingertip capillary sampling in the 8-11 y boys. The preliminary investigation, established that GLP-$1_{7\text{-}36}$ and glucose could confidently be measured in 8-11 y boys by fingertip capillary sampling. The measurement of glucagon, leptin and insulin as well as GLP-$1_{7\text{-}36}$ and glucose during active and seated video could have proved interesting in view of the association these peptides have to obesity (Guran et al., 2008; Woods et al., 2006). The investigation of satiety-related signals in response to active and seated video gaming in 8-11 y boys, in Chapter V therefore, was restricted to GLP-$1_{7\text{-}36}$ and glucose. The methodological limitations of the measurement of the satiety-related signals in 8-11 y boys will thus focus on GLP-$1_{7\text{-}36}$ and glucose.

Research in adult populations suggests that the levels and responses of GLP-$1_{7\text{-}36}$ to EI may be reduced due to obesity (Holst, 2013). Despite the evidence that GLP-$1_{7\text{-}36}$ appears to become dysregulated in adult obesity, the present study population which was mainly lean boys, was not split by BMI classification. To date there is very little evidence to suggest that GLP-$1_{7\text{-}36}$ levels and responses are affected in relation to body mass, in children. Indeed in terms of fasting concentrations of GLP-$1_{7\text{-}36}$, these have been reported to be similar in lean, overweight and obese children (Horner & Lee, 2015; Lomenick, White, Smart, Clasey, & Anderson, 2009; Reinehr, De Sousa, & Roth, 2007). There is nonetheless, a small amount of evidence that post-prandial levels of the aforesaid hormone are lower in obese children, than in those classified as lean (Horner & Lee, 2015; Tomasik, Sztefko, & Malek, 2002). Furthermore, in response to exercise, there is little knowledge regarding plasma levels of GLP-$1_{7\text{-}36}$ between lean and overweight children. Although similar to feeding studies, the responses of 15 y males were found to be 25% lower in those classified as overweight, in comparison to those of lean body mass (Chanoine et al., 2008). Indeed this may indicate that low concentrations of GLP-$1_{7\text{-}36}$ could also facilitate child obesity (Chanoine et al., 2008; Horner & Lee, 2015).

Similar to body mass, sex and age differences in relation to the responses of GLP-1_{7-36} have not been widely explored in paediatric populations. Fasting concentrations of GLP-1_{7-36}, have been found to be similar in 10-12 y boys and girls (Horner & Lee, 2015; Reinehr, De Sousa & Roth, 2007). In addition, in the same study population, fasting GLP-1_{7-36} was not found to be related to age (Horner & Lee, 2015; Reinehr, De Sousa & Roth, 2007). A reason for such a lack of studies of GLP-1_{7-36} and indeed other appetite-related signals in paediatric groups may be that measurements have been precluded by blood sampling techniques. The usual method of blood sampling in adults is the antecubital venous technique. The aforementioned method was deemed too invasive for paediatric populations, particularly for the 8-11 y male participants in the present research. To overcome this, the novel method of fingertip sampling was utilised. To the best of our knowledge, this is the first investigation to examine GLP-1_{7-36} in response to both active and seated video gaming in a paediatric population, which is not only novel, but an asset to the thesis.

In relation to glucose, there is evidence of impaired homeostatic levels in children due to obesity (Bedogni et al., 2012) which suggests that the present study population should have been split by BMI classification. In spite of the aforementioned evidence however, the levels of glucose observed during both active and seated video gaming in Chapter V (Figure 5.6b) displayed no evidence of impairment. Indeed, during both gaming interventions when food and fluids were offered *ad-libitum*, blood glucose remained within the normal range for the boys, even when a greater amount of CHO was consumed during active video gaming. Nonetheless, comparison of glucose responses to active video gaming between lean, overweight and obese children does warrant further research.

6.4 Future directions for research and practice

6.4.1 Implications for research

The 'Active Gaming Questionnaire' informed the design of the studies described in Chapters IV and V and emphasised the need to establish active video gaming practices in a study population, prior to examination of acute appetite, EI and PA. The reason for this is that there will likely be differences in real-life active gaming practices, according to age and sex. In addition, there appears to be on-going advances in active video gaming technology and some major developments and improvements have been made during the course of the present research. In view of this, future investigations should be encouraged to establish the up to date, real-life active video gaming practices in relation to the study population. In doing so, a more accurate and clear representation of appetite, EI and PA responses to active video gaming would be obtained which could then more accurately inform paediatric energy balance research and childhood obesity prevention.

Active video gaming paediatric appetite research is relatively novel and to date, there is no evidence that subjective or objective appetite responses to gaming EE differ according to body mass. Presently, no differences were found in the subjective appetite sensations of hunger, prospective food consumption and fullness. In relation to the objective measurement of appetite-related satiety signals, there was a significant increase in blood glucose and a slight but non-significant rise in plasma GLP-$1_{7\text{-}36}$ in response to active video gaming. Future research would therefore benefit from the direct comparison of subjective and objective appetite responses between lean, overweight and obese paediatric groups.

The present thesis was focussed on the acute appetite, EI and PA and thus relative energy balance in response to active video gaming, in 8-11 y boys. Thus far only one previous active video gaming investigation with adolescent males has been able to extend the period of measurement to 3 days (Gribbon et al., 2015). The cited study established that EE increased due to active video gaming and so following a test meal and up to 24 h later, the participants were in energy balance. Subsequently however, there was compensation by a down-regulation in EE. Presently, appetite, EI and PA responses and the relative energy balance of the boys could only be monitored up to 1 h post active video gaming. As no compensation occurred for the increased PA and the EI during game play, in the test meal, it would be beneficial, to extend the period of monitoring to determine if and when compensation takes place. However, the difficulties encountered when recruiting and working with 8-11 y boys outlined in section 6.3.1, should be borne in mind when planning such studies. It would be more challenging for researchers to track subjective and objective appetite, PA, EE and would place greater demands on participants, their parents and schools. When funds and expertise are available however, the measurement of EE and subsequent energy balance by DLW would remove some of the challenges for researchers and burden on participants thus could advance future work considerably.

The fingertip blood sampling procedure utilised was an asset to the present work however, only blood glucose and plasma GLP-$1_{7\text{-}36}$ were able to be accurately measured in the 8-11 y boys. The objective investigation of appetite would be further advanced if this less invasive technique could be used to accurately measure hormones related to both hunger and satiety in the fasted, post-prandial and post-absorptive states.

6.4.2 Implications for practitioners and parents

With regards to eating and drinking during active and seated video game play, the present research demonstrated that EI exceeded the PA elicited and the subsequent estimates of EE, resulted in a positive relative energy balance state in the 8-11 y boys. The examination of the foods and fluids consumed by the boys during active and seated video gaming revealed that crisps were the main contributors to the positive relative energy balance. The lack of differences in subjective and objective appetite responses, apart from for blood glucose, also suggests that food and fluid intake during active and seated video gaming could be largely controlled by hedonic rather than hormonal mechanisms. The implications of these findings for parents are that they should discourage their children from consuming unhealthy, energy dense foods and drinks during both seated and active video game play. If this specific eating behaviour were to become habitual and continually exceed active gaming EE, it has the potential to lead to long term positive energy balance and so contribute to increased weight status in childhood and possible obesity.

For the majority of time during the 90 min of active video game play, the PA intensity levels attained by the boys were light compared with sedentary levels of activity when seated video gaming. Although the active video game play of 8-11 y boys with Nintendo Wii ™ does not contribute to MVPA, it will be beneficial in terms of reducing sedentary time. In contrast, seated video gaming increased sedentary time and is a behaviour believed to be as detrimental to health as physical inactivity. Parents should therefore, encourage their children to play active video games rather than those that are seated. In addition, the Government guidelines for PA should incorporate a recommendation for time (min) spent participating in sedentary activities, such as seated video game play, as has already been done in Australia and Canada (Australian Government. Department of Health, 2014; Tremblay et al., 2012). The recommendations should also include examples of the typical sedentary activities during childhood, as this could help parents to recognize them and thus where necessary take action to reduce sedentary behaviour in their children.

6.5 Main conclusions

Based on the findings of the studies conducted within the present thesis, the following conclusions can be made;

- Children typically play the Nintendo Wii™ and Nintendo Wii™ Sports, once or twice per week for 81±50 min. The majority of the children, consumed food and (or) fluids during active video game play. The most commonly consumed foods and fluids were fruit, crisps, fruit squash and milk.
- When foods (fruit and crisps) and fluids (fruit squash and milk) were offered *ad-libitum* during video game play, active video gaming reduced the EI of the 8-11 y boys, by an average of 10% of daily EAR for energy, in comparison to seated video gaming. When estimated active video gaming EE was deducted from EI however, the relative energy balance of the boys remained positive.
- Subjective sensations of hunger, prospective food consumption and fullness were no different between active and seated video gaming and so did not provide explanation for the reduced EI during active video game play.
- The objective measurement of metabolic satiety-related signals by fingertip capillary sampling during game play revealed that blood glucose concentrations were greater during active video gaming. The inspection of macronutrient intakes, established that the greater blood glucose concentrations appeared to be due to the larger amount of CHO, in proportion to percentage total EI, ingested during active video gaming. The larger amount of CHO was caused by the high intake of crisps.
- Although not significant, the plasma concentrations of GLP-$1_{7\text{-}36}$ showed a greater increase from fasting levels, during active video gaming, which could indicate a satiety response. The

proposed satiety response however, was not strong enough to suppress EI during active video gaming. Therefore, the boys were still able to consume 21% of total daily EAR for energy which resulted in a positive relative energy balance (0.99 MJ).

- Similar to seated video gaming, there was no compensation for the EI during active video game play, in the post-gaming test meal 1 h later. Instead, the relative energy balance increased to +2.06 MJ, although this was still considerably lower than for seated video gaming (+3.34 MJ).
- The positive energy balance state at cessation of game play and the greater concentrations of plasma GLP-$1_{7\text{-}36}$, could indicate that EI was largely driven by hedonic mechanisms which appear to have the capacity to supersede homeostatic satiety-related signals. Therefore, children who habitually eat and drink during active video gaming especially, snacks such as crisps which are energy dense snacks and of low micronutrient value, may be at risk of longer term positive energy balance which could lead to overweight or obesity.
- For the majority of time during the 90 min of active video game play with Nintendo Wii™ Sports tennis (~ 70%), the boys elicited only light intensity PA. Consequently, active video gaming does not help to meet the recommendation that children should undertake at least 60 min of MVPA, per day. However, the light PA induced by active video gaming will contribute to a reduction in sedentary time.

CHAPTER VII

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CHAPTER VIII

APPENDICES

northumbria
UNIVERSITY NEWCASTLE

The Newcastle Children's Active Gaming Study

ACTIVE GAMING QUESTIONNAIRE

Active video games are new generation computer games. The purpose of this questionnaire is to find out whether 7-to-11 year old children from Newcastle upon Tyne play active video games and if so to learn about the patterns of play.

We would like to invite your child and you as their main carer to complete the questionnaire together. The reason for this is that you will find it is more appropriate for some of the questions to be answered primarily by your child and others primarily by you. When the questions are more appropriately answered by your child please give them the freedom to answer as they wish.

As a guide for you when answering the questions, please refer to the following explanations of active gaming consoles and active video games.

Active gaming consoles are the gaming consoles that enable the physical interaction needed to play certain types of games by tracking the body movement of the player. A few examples of active gaming consoles are;

- Nintendo Wii
- XBOX 360 Kinect
- Sony Play Station Move
- Sony Eye Toy: Kinetic

Active video games are the games that can be played on active gaming consoles. They replicate sports for example athletics and football, or fun physical activities such as rafting, flying and dance. To play an active video game the player must physically interact with the console by moving their body sometimes with a device known as a controller. A few examples of active video games are;

- Wii Sports
- Kinect Adventures
- Just Dance 3

Active video games do not include the games that are played whilst sitting down such as U Draw Studio: Instant Artist, Bionicle Heroes or LEGO: Harry Potter.

SECTION 1: YOUR CHILD AND ACTIVE GAMING

1. Would you please tell us the name of the Primary School in Newcastle upon Tyne your child currently attends?

[ ]

2. Is your child a boy or girl?

❑ Boy

❑ Girl

3. Would you please tell us the exact age of your child?

____________________ Years

____________________ Months

4. Does your child have access to play an active gaming console?

❑ Yes

❑ No

If you answered 'Yes' please continue to question 5. If you answered 'No' please now go to section 2.

5. Would you please tell us where your child has access to play the active gaming console?

Please tick all that apply.

❑ At home

❑ At a friend's home

❑ At a relative's home

❑ Other- please specify: [ ]

6. Would you please tell us which active gaming consoles your child plays?

Please tick all that apply.

❑ Sony Play Station Move ❑ XBOX 360 Kinect

❑ Nintendo Wii ❑ Sony Eye Toy: Kinetic

❑ Other – please specify:

7. There are many **active video games**, a few examples are; Wii Sports, Kinect Sports or Zumba Fitness. Does your child play **active video games** on the active gaming console?

Please tick most appropriate answer.

❑ No

❑ Yes

❑ Don't know

If you answered 'no' or 'don't know', please now go to section 2. If you answered 'yes', please continue to question 8.

8. Would you please write down the names of your child's favourite **active video games** (up to a maximum of three games)?

9. Would you please tell us the name of the **active video game** that your child plays most often?

10a. During the last 7 days, on how many days did your child play **active video games**?

Please tick most appropriate answer.

❑ Never ❑ 1 to 2 days

❑ 3 to 4 days ❑ 5 to 6 days

❑ Everyday

10b. When your child does play how much time do they usually spend playing **active video games**?

________________________ Hours

________________________ Minutes

11. On which day of the week does your child tend to play **active video games** most often?

[ ]

12a. Does your child play **active video games** with other people (e.g. their friends or relatives)?

❑ Yes

❑ No

12b. If you answered **'Yes',** please tell us who they play **active video games** with most frequently.

Please tick one answer only.

❑ Friend/friends ❑ Brother/s and/or sister/s

❑ Parent/parents ❑ Cousin/s

❑ Grandparent/s ❑ Aunt or Uncle

❑ Child-minder

❑ Other – please specify: [ ]

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13. Please tell us the main reason why your child plays **active video games**?

[ ]

14a. Does your child tend to eat and/or drink whilst they play **active video games**?

❑ Yes

❑ No

14b. If you answered **'yes'**, in the space provided would you please tell us the types of food or drink they eat or drink most often whilst they play **active video games**?

15a. Do you consider **active video games** to be an alternative form of exercise for your child?

❑ Yes

❑ No

❑ Don't know

15b. If you answered **'Yes'** or **'No'** please give a reason for your answer.

SECTION 2: ABOUT YOU AND YOUR FAMILY

1. To which of these ethnic groups do you consider your child belongs?

❑ White

❑ Black

❑ Asian

❑ Mixed

❑ Other

2. Would you please tell us how many other children there are in your family?

3. As your child’s main carer which of these most accurately describes your relationship to your child?

❑ Mother

❑ Father

❑ Grandparent

❑ Other- please specify:

4. Would you please tell us which of these most appropriately describes your marital status?

❑ Single

❑ Married/Co-habiting

❑ Divorced/Separated

❑ Widowed

5. Would you please tell us the highest qualification achieved within your household?

6. Would you please tell us who provides the main source of income for your family?

❑ Child’s mother

❑ Child’s father

❑ Benefits

❑ Other

7. What is the main provider’s present employment status?

❑ Working full-time

❑ Working part-time

❑ Studying (in full or part-time education)

❑ Not working at present

8. If the main provider is working full or part-time, would you please tell us their occupation?

9. What is the approximate annual income for your household before tax or any other contributions are deducted?

- ❑ Up to £14,999
- ❑ £15,000-£19,999
- ❑ £20,000-£29,999
- ❑ £30,000-£49,999
- ❑ £50,000-£99,999
- ❑ £100,000 and over

10. If at some future date we wanted to invite your child to take part in a further active gaming study, may we contact you to see if you would be willing to help us again?

- ❑ Yes
- ❑ No

Questionnaire number	

Thank you!

The Newcastle Children's Active Gaming Study

We really appreciate the time you have taken to complete this questionnaire! If you have any questions please contact Susan Allsop at the School of Life Sciences, Northumbria University, Northumberland Building, Northumberland Road, Newcastle upon Tyne NE1 8ST either by telephone on 0191 243 7018/ 07884 025031 or by emailing s.allsop@northumbria.ac.uk.

APPENDIX B: Recruitment letter to school

Susan Allsop
Department of Sport, Exercise & Rehabilitation,
Faculty of Health and Life Sciences,
Northumberland Building,
Northumberland Road,
Newcastle-upon-Tyne.
NE1 8ST
Direct Line: 0191 2437018
Email: s.allsop@northumbria.ac.uk

DATE

Dear (Head Teacher name),

My name is Susan Allsop and I am a student at Northumbria University currently studying a PhD investigating 'Food intake and hormonal appetite responses to seated and active gaming in 8-to-11 year-old boys.' The reason for the study is that children aged ≤ 11 years have been found to commonly consume foods and drinks during seated media activities such as television viewing and computer game play. Such food and drink intake during these activities is thought to contribute to childhood overweight. During recent seated and active gaming trials, no differences were found in the food intake and appetite ratings of 8-to-11 year-old boys. Consequently active games might have the same effects on childhood weight as television and computer gaming. Further in-depth investigation of food intake and hormonal appetite responses during seated and active gaming would therefore be beneficial to child health.

The study has received full ethical clearance from the Ethics Committee within the Faculty of Health and Life Sciences, Northumbria University and all members of the research team have enhanced CRB clearance. I am therefore writing to ask if you would give permission for your school to assist me with the research project. If you are happy to give your permission, I will merely ask your school to be a venue for recruitment and a small amount of data collection. I would like to assure you that the time-demands of the study upon the school, staff and pupils will be very low.

Over the past few years I have been involved in various research projects that have collaborated with local primary schools. The schools, staff and pupils have generally found the research experience to be positive and gain a great amount of enjoyment from taking part. To show appreciation for any help your school is able to provide with my current project, I would be happy to provide a fun physical activity session to your pupils.

Thank you for taking the time to read this letter and I will contact you in the next few days to provide further details of the project and discuss the possibility of your school's participation. In the meantime, if you have any questions or would like to know more about my research project, please do not hesitate to contact me on the details provided above.

Yours Faithfully,

Susan Allsop (BSc Hons)

APPENDIX C: Recruitment letter to parents

northumbria UNIVERSITY NEWCASTLE
Faculty of Health & Life Sciences

Susan Allsop,
Department of Sport, Exercise & Rehabilitation,
Faculty of Health and Life Sciences,
Northumberland Building,
Northumberland Road,
Newcastle-upon-Tyne.
NE1 8ST
Direct Line: 0191 2437018
Email: s.allsop@northumbria.ac.uk

Dear Parent/Guardian,

My name is Susan Allsop and I am a PhD student at Northumbria University. I am currently investigating the 'Food intake and hormonal appetite responses to seated and active gaming, in 8-to-11 year-old boys.' The reason for the study is that it is commonplace for children aged ≤ 11 years to eat and drink during seated media activities such as television viewing and computer game play. Eating and drinking during these activities is thought to contribute to childhood overweight. In a recent study no differences were found in the food intake and appetite ratings of 8-to-11 year-old boys when they played either an active video game or a seated computer game. Active games might therefore have the same effects on childhood weight as television and computer gaming. Further in-depth investigation of food intake and hormonal appetite responses during seated and active gaming would therefore be extremely beneficial to child health.

I would be very grateful therefore if you would consider allowing your child to take part in my research. If you are both interested please read the details of the project outlined in full in the two information sheets I have enclosed with this letter. Should your child be happy to participate, please both sign the enclosed ***informed consent forms*** and return them in the envelope provided, to their class teacher no later than the (agreed date).

Whilst you are considering my invitation, I would like to make you aware that there is no obligation for your child to take part and there will be no disadvantage to them should they decide not to. I would also like to inform you that all members of the research team have enhanced CRB clearance.

Thank you for taking the time to read this letter. If you have any further questions or would like to know more about my research project, please do not hesitate to contact me on the details provided above.

Yours Faithfully,

Susan Allsop (BSc Hons).

northumbria UNIVERSITY NEWCASTLE
Faculty of Health & Life Sciences

CHILD INFORMATION

HELLO!

I would like to invite you and your friends to help me out with some research by playing some video games on Nintendo Wii™ and Nintendo 3DS™. If you would like to take part I will come along to meet you and your friends and tell you all about my research. I will then measure how tall you are and how much you weigh. You will also be asked to answer a few questions, which I will help you with.

Over the next four weeks, I will come to your school again and visit you and your friends and spend one day with you each week. On each visit I will ask you to play games on one of the gaming consoles for 90 minutes. The games will either be Nintendo Wii™ Sports or Mario and Sonic at the London 2012 Olympics games. While you play the games I will measure how much movement you make and ask you some questions about your appetite. On two of the days while you are playing the games you will be able to eat or drink if you want to.

It would be great if you think you can help me with this. If you would like to, all you need to do is tell one of your parents or your carer.

Don't worry if you can't take part or you don't want to. If you start the study and you don't like it and want to drop out then that is fine too.

Thank you, for reading this. I really hope you would like to take part!

Susan.

Faculty of Health & Life Sciences

ACTIVE GAMING STUDY INFORMATION FOR PARENT OR GUARDIAN

I am sure you would like to know more about the study so the purpose of this information sheet is to provide you with sufficient information so that you can then give your informed consent. It is very important that you read this document carefully, and raise any issues that you do not understand with the principal investigator.

The purpose of the study:
The purpose of the study is to find out if there are any differences in the food intake and hormonal appetite responses to active video gaming compared with seated computer gaming in 8-to-11 year-old boys. To then explore their awareness of these responses.

Why has my child been selected to take part?
The study is a continuation of previous research with 8-to-11 year-old boys that investigated the effects of active gaming on food intake and appetite ratings. Your child has been invited to take part because he is male, attends a primary school in Newcastle-upon-Tyne and is within the age-group being studied which is 8-to-11-years.

What will my child be asked to do?
The research team will visit your child's school on an agreed date. You and your child will be invited to meet us so we can give a full explanation of how the study and gaming sessions will be run. Most importantly it will be an opportunity to give a demonstration of how the blood samples will be taken. Your child will also be able to play and become familiar with the gaming consoles and games.

At this time, height, seated height, weight and waist circumference will be measured, in private and in the presence of two researchers. Your child will be asked to wear their usual school PE clothing for this. A food preference questionnaire and an eating behaviour questionnaire will also. The latter has been used comprehensively in child studies to assess whether food intake is consciously restricted.

Gaming Sessions: Venue
Please refer to the enclosed diagram of the study. For the gaming sessions your child will be asked to visit the Northumbria University Sport and Exercise Laboratories. The sessions will be on two separate days from 9:00am until 13:00pm, in consecutive weeks. Your child and one other boy will be escorted by two members of the research team and transported by University bus from school to the Sport and Exercise Laboratories.

Gaming Sessions: Preparation
As food can affect appetite and energy levels, it is important that your child eats the same foods and drinks in the same amounts from 5:00pm the day before each visit. You and your child will be asked to complete a food diary from 5:00pm the evening before each gaming session until arrival at school the next morning. You and your child will be asked to weigh all food and drink items they consume and record these in the diary. A food diary and food-weighing scales will be provided. It would also be helpful if your child tried to avoid participating in physical activity on these days, as this can affect results. Researchers will liaise with the staff at the school to ensure exemption from PE and drama lessons.

Gaming Sessions
Your child will be asked to play one of the following games each week for 90 minutes during which food and drink items will be offered to them;

- Mario and Sonic at the London 2012 Olympic Games on Nintendo 3DS™;
- Nintendo Wii™ Sports-Tennis on Nintendo Wii™.

Gaming Session: Measuring Appetite
Appetite will be measured in two ways. Firstly, with visual analogue scales which ask your child to rate their appetite.

Secondly, it is measured from particular hormones that circulate in the blood. For this, a small amount of blood will need to be taken. A fully trained researcher will do this by pricking your child's finger with a device called a lancet and collecting it into a small test-tube. A procedure that has been risk assessed. One sample will be taken at three stages during each of the two gaming sessions; at the beginning, half-way through (at 45 minutes) and at the end (90 minutes), as shown in the study diagram.

Resting Session
Immediately following each gaming session, your child and will be asked to rest for 1 hour.

Post-Gaming Meal
Following the resting session, the research team will provide your child with lunch consisting of their preferred foods and drinks (established from the food preference questionnaire). During the lunch break, the researcher will conduct a dietary recall interview with your child.

Are there any reasons why my child should not take part?
Your child will be excluded from the study if they have any medical conditions that will affect their participation or any allergies or intolerances to any foods provided in the study. They could also be excluded if they are unable to take part on any of the study days due to illness which causes their absence from school.

Will my child's participation involve any physical discomfort?
The process of finger lancing has been risk-assessed but it can make the fingertip feel a little uncomfortable and sensitive and may produce slight bruising. To try to avoid any discomfort, your child will be asked if they have a preferred finger from which the blood sample will be drawn. In order to help minimise the occurrence of any discomfort, proper aseptic methods will be followed, and all sampling shall be conducted by professionally trained researchers.

Will my child's participation involve any psychological discomfort or embarrassment?
No.

How will confidentiality be assured?
When processing your child's personal data, the principal researcher will ensure compliance with current data protection legislation (Data Protection Act, 1988). All of the data collected will have an identification number in place of your child's name to prevent association between participant and all study documents. Consent forms containing both of your names will be stored separately from those containing identification numbers and locked in a filing cabinet. All documents containing information about your child will be stored in a locked filing cabinet to be accessed only by the principal researcher and used only for the originally intended purpose**.** All electronic data will be stored on password-protected computers.

Who will have access to the information that my child and I provide?
Any information and data gathered during this research study will be available only to the principal investigator (Susan Allsop) and the PhD supervisory team. Should the research be presented or published in any form, it will be generalized so that your child's personal information and data will not be identifiable.

How will my child's information be stored or used in the future?
All information and data collected from your child during the research will be stored in line with the Data Protection Act and destroyed 5 years following completion of the PhD programme. The information provided from you and your child will be used by the principal researcher (Susan Allsop) only, for the purpose of this study. The data may be presented at conferences and also published in peer reviewed journals and at no point will your child's identity be associated with the results.

Has the investigation received appropriate ethical clearance?
Yes, the study and its protocol have received full ethical approval from the Faculty of Health and Life Sciences Ethics Committee. If you require confirmation of this please contact the Chair of this committee, stating the title of the research project and the name of the principal investigator;

Chair of the Faculty of Health and Life Science Ethics Committee (Dr Les Ansley)
Northumberland Building
Northumbria University
Newcastle upon Tyne
NE1 8ST.
Email: les.ansley@northumbria.ac.uk

Will my child and I receive any financial rewards or travel expenses for taking part?
No, but your child will receive a certificate thanking them for completing the two gaming sessions.

How can my child withdraw from the project?
You and your child are reminded of your right to withdraw from the study at any time without prejudice. If you or your child chooses to do so, please email the principal investigator (email: s.allsop@northumbria.ac.uk) as soon as possible to enable the withdrawal and discuss how you would like your child's data to be dealt with. After your child has completed the two gaming sessions, his data can still be withdrawn by contacting the principal researcher (their contact details are provided below). Please note however that it might not be possible to withdraw your child's data if it has already been analysed or published, so please contact the investigator within one month of the fourth gaming session if you do wish to withdraw his data. However the research that you and your child are being invited to take part in will be more representative and valuable if fewer people withdraw, so please discuss any concerns you might have with the principal researcher (Susan Allsop).

If I require further information who should I contact and how?
Thank You for taking the time to read this information leaflet. Please do not hesitate to contact me (Susan Allsop – Principal Investigator) using the following contact details and quoting the participant number shown.

Department of Sport and Exercise Sciences
Faculty of Health and Life Sciences
Room 431, Northumberland Building
Northumbria University
NE1 8ST
Tel: +44(0)191 243 7018
Mob: 07884 025031
Email: s.allsop@northumbria.ac.uk

Participant number:

APPENDIX E: Parent and child consent forms and tissue consent

northumbria
UNIVERSITY NEWCASTLE
Faculty of Health & Life Sciences

INFORMED CONSENT FORM PARENT/GUARDIAN & CHILD

Project Title:

Principal Investigator: Susan Allsop

Participant Number: ______

please tick where applicable

My child and I have read and understood the Participant Information Sheet. ☐

My child and I have both had an opportunity to ask questions and discuss this study and have received satisfactory answers. ☐

My child and I understand he is free to withdraw from the study at any time, without having to give a reason for withdrawing, and without prejudice. ☐

I agree for my child to take part in this study. ☐

My child and I would like to receive general feedback on the overall results of the study at the email address given below. We understand that we will not receive individual feedback. ☐

Email address……………………………………………………………………

Signature of parent/guardian participant...

Date....…………………..

NAME IN BLOCK LETTERS...………………………………………….

TELEPHONE: Home………………………….. Work……………………….. Mobile……………………

Signature of child participant...

Date..........................

NAME IN BLOCK LETTERS...………………………………………….

SCHOOL CLASS and YEAR..………………………………………….

Signature of researcher..

Date....…………………………

NAME IN BLOCK LETTERS...………………………

northumbria UNIVERSITY NEWCASTLE
Faculty of Health & Life Sciences

INFORMED CONSENT FORM PARENT/GUARDIAN & CHILD

FOR USE WHEN TISSUE IS BEING REMOVED BUT NOT STORED

Project Title: The determination of acute energy intake and metabolic appetite responses to habitual active gaming in 8-to-11 year-old boys.

Principal Investigator: Susan Allsop

Participant Number: __________

I agree that the following tissue or other bodily material may be taken and used for the study:

Tissue/Bodily material	Purpose	Removal Method
Capillary blood plasma	*For analysis of glucose.*	*Via finger-prick*

I understand that if the material is required for use in any other way then that will be explained to me and my consent to this will be specifically sought. I understand that I will not receive specific feedback from any assessment conducted on my samples, but should any kind of abnormality be discovered then the investigator will contact me.

Signature of participant... Date..........................

Signature of Parent / Guardian in the case of a minor

.... Date..........................

Signature of researcher... Date..........................

APPENDIX F: Dutch eating behaviour questionnaire-child

Dutch Eating Behaviour Questionnaire for Children **Scoring sheet**

Instructions
Below you'll find 20 questions about eating.
Please read each question carefully and tick the answer that suits you best.
Only one answer is allowed. Don't skip any answer.
There are no incorrect answers; it's **your opinion** that counts.

1.	Do you feel like eating whenever you see or smell good food?	No	Sometimes	Yes
2.	If you feel depressed do you get a desire for food?	No	Sometimes	Yes
3.	If you feel lonely do you get a desire for food?	No	Sometimes	Yes
4.	Do you keep an eye on exactly what you eat?	No	Sometimes	Yes
5.	Does walking past a candy store make you feel like eating?	No	Sometimes	Yes
6.	Do you intentionally eat food that helps you lose weight?	No	Sometimes	Yes
7.	Does watching others eat make you feel like eating too?	No	Sometimes	Yes
8.	If you have eaten too much do you eat less than usual the next day?	No	Sometimes	Yes
9.	Does worrying make you feel like eating?	No	Sometimes	Yes
10.	Do you find it difficult to stay away from delicious food?	No	Sometimes	Yes
11.	Do you intentionally eat less to avoid gaining weight?	No	Sometimes	Yes
12.	If things go wrong do you get a desire for food?	No	Sometimes	Yes
13.	Do you feel like eating when you walk past a snack bar or fish and chips stand?	No	Sometimes	Yes
14.	Have you ever tried not to eat in between meals to lose weight?	No	Sometimes	Yes
15.	Do you have a desire to eat when you feel restless?	No	Sometimes	Yes
16.	Have you ever tried to avoid eating after your evening meal to lose weight?	No	Sometimes	Yes
17.	Do you have a desire for food when you are afraid?	No	Sometimes	Yes
18.	Do you ever think that food will be fattening or slimming when you eat?	No	Sometimes	Yes
19.	If you feel sorry do you feel like eating?	No	Sometimes	Yes
20.	If somebody prepares food do you get an appetite?	No	Sometimes	Yes

PLEASE CHECK, TO BE SURE THAT YOU TICKED EVERY QUESTION.

APPENDIX G: Visual analogue scales for appetite

WEEK no. : Start of gaming (3:30pm)

1. **How hungry do you feel now?**

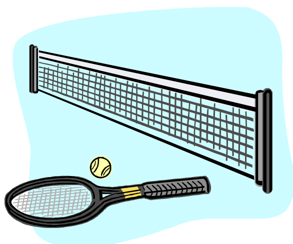
Very hungry — Not at all hungry

2. **How much would you like to eat now?**

A lot — Nothing at all

3. **How full do you feel now?**

Very full — Not full at all



APPENDIX H : Food diary

FOOD DIARY

Weighing your child's food:

Firstly, we would like to thank you for agreeing to take part in this study and for helping your child to weigh and record their food and drink intake. We have provided an outline of how to record everything your child eats and drinks from 5:00pm the evening before until their arrival at school on the day of active gaming club. Should you have any queries on how to complete the food diary please do not hesitate to contact me on 0191 2437018.

Day and Date

Please write down the date at the top of the page on the day of recording.

Time and Meal

In the column provided please note the time of each eating occasion and also the type of meal consumed i.e. whether it is breakfast, lunch, dinner, supper or a snack.

Description of Food

In the column provided please describe the food your child eats in as much detail as possible. Be as specific as you can with regards to **cooking methods** (fried, grilled, baked etc.) and any **additions** (butter/margarine, sugar/sweeteners, sauces, pepper etc.).

Homemade dishes:

If your child eats any homemade dishes e.g. chicken casserole, please record the name of the recipe, ingredients with amounts (including water or other fluids) or the whole recipe, the number of people the recipe serves and the cooking method. Write this down in the recipe section. Record how much of the whole recipe your child has eaten in the portion size column.

Take-aways and eating out:

If your child has eaten **take-aways** or **dishes not prepared at home** such as at a restaurant or a friend's house, please record as much detail about the ingredients as you can e.g. vegetable curry containing chickpeas, aubergine, onion and tomato.

Amount Consumed

In the 'weight before' and 'weight after' columns the size of the portion of food eatencan be described using:

weights from labels e.g. 4oz steak, 420g tin of baked beans, 125g pot of yoghurt
weighing/measuring food and drink items using scales/measuring jugs.

We would like to know the **amount that your child actually ate** which means taking leftovers into account. You can do this in two ways:
Record what was served and note what was not eaten e.g. 3 tbsp of peas, only 2 tbsp eaten; 1 Weetabix ate ½.
Only record the amount actually eaten i.e. 2 tbsp of peas; ½ Weetabix.

Brand Name

Please note the **brand name** (if known). Most packaged foods will display the brand name e.g. Bird's Eye, Hovis or supermarket own brands.

CHECKLIST		
Food/Drink	**Description & Preparation**	**Amount (remember you can write the g, ml or kg values if you have them!)**
Bacon	Lean or streaky? Fried or grilled rashers?	Number
Baked Beans	Normal or reduced sugar/salt?	Tablespoons, tin or carton size
Beefburger/Hamburger	Home-made, packet or take-way? Fried, micro-waved or grilled? With bread roll?	Number
Biscuits	Plain, chocolate, sweet, crisp bread, cheese, wafer, home-made? What brand?	Number
Bread	Wholemeal, granary, white, multigrain? Currant, fruit, malt? Large or small loaf? Thick, medium or thin slices? Brand?	Number of slices
Bread Rolls	Wholemeal, granary, white? Size? Crusty or soft? Brand? If with filling remember to record it?	Number of rolls
Breakfast Cereal	What sort? Cornflakes, Weetabix etc. What brand?	Number, weight or tablespoons
Bun	What sort: Iced, currant, sweet or plain? Large or small? Brand?	Number
Butter	Ordinary or low fat spread? Brand?	Spread: thickly, average or thinly
Cake-small and large	What sort: Cream, iced, chocolate etc.? Brand?	Number, weight or slices
Cheese	What sort: hard, soft, spread, cream, low-fat, mature, mild? Brand?	Tablespoons
Chips	Frozen, oven, microwave, crinkle-cut, chip-shop, MacDonald's etc? How cooked? In what oil? Brand?	Tablespoons or weight
Chocolate	What sort: Milk, white, plain? Name and Brand?	Number or weight of bar
Chops	What sort: lamb or pork? Lean or fatty? Large and small? Fried, grilled, baked	Number or weight
Coffee	Include milk & sugar! Skimmed, semi, whole milk?	Small, medium or large cup or mug. How much milk/sugar?

Cooking oil	Type, Brand?	How many teaspoons/tablespoons?
Cream	Half, whipping, single, double, clotted, low-fat, fresh or substitute?	How many tablespoons?
Crisps	Brand name? Normal, low-fat or low-salt?	Packet weight
Egg	How was it cooked: boiled, fried, scrambled, poached, omelette etc.	Number and size
Fish	What sort: Fried, boiled, grilled, poached, micro-waved? Pickled, smoked, salted? Batter, breadcrumbs? Tinned with oil or tomato sauce? Size? Brand where possible?	Weight.
Fish fingers/cakes	What sort; large, medium or small? Fried or grilled? Brand?	Number
Fruit–fresh	What type (e.g. banana, apple and orange)? Brand (e.g. Bramley, Golden Delicious)?	Number
Fruit-canned/stewed	In fruit juice or syrup? Type of fruit? With or without sugar?	Tablespoons or tin size
Fruit-juice	What sort; sweetened or unsweetened? Brand?	Glasses or cups, small, medium or large
Gravy	Thick or thin? Instant, packet or homemade?	Tablespoons
Honey	Brand? Clear?	Teaspoons
Ice-cream	Dairy or non-dairy? Flavour, variety? Brand?	Tablespoons
Jam	Brand? Normal or low-sugar?	Teaspoons
Kidney	Fried or stewed? Pig, lamb or ox?	Weight.
Liver	Fried or stewed? Pig, lamb or ox?	Weight.
Margarine	Soft, hard? Polyunsaturated, low-fat, very low-fat? Brand?	Spread thickly, medium or thinly or number of tablespoons/teaspoons.
Marmalade	Brand? Normal or low-sugar?	Spread thickly, medium or thinly or number of tablespoons/teaspoons.
Mayonnaise	Brand? Normal or low-fat?	Teaspoons or tablespoons.

Meat	What sort: lean or fatty? Fried, grilled, roast, BBQ, micro-waved etc.? Any gravy? If so see GRAVY on checklist!	Slices or weight.
Milk	Full cream, semi-skimmed, skimmed? Sterilised, UHT, flavoured, powdered, soya?	
Mince	Beef, lamb, pork turkey? On it's own, with vegetables, gravy (see VEGETABLES and GRAVY on checklist)? Fatty or lean? Brand?	Tablespoons or weight.
Pasta, Spaghetti	Canned, fresh or boiled? White or wholemeal? In sauce (see SAUCE on checklist) ? Brand?	Tablespoons or weight.
Pie, Pasty, Pastry	What sort: meat, vegetable, fruit etc..? Individual or a slice? What type of pastry? Brand?	Number or weight
Peanuts	Ordinary, salted or dry roasted?	Packet weight
Porridge	How made: with all milk/ milk and water/ cream? Type of milk (see MILK on checklist)? With sugar or honey? Brand?	Small, medium or large bowl
Potatoes	Baked, boiled (with or without skin?), mashed, creamed, fried, chips (see CHIPS on checklist), roast, instant? With butter, margarine etc., (see BUTTER or MARGARINE on checklist)?	Tablespoons or weight.
Pudding	What sort and brand? Is it jelly, mousse, milk pudding, sponge etc? If with cream, see CREAM on checklist.	Tablespoons or slices.
Rice	Brown or white? Boiled or fried? Brand? If rice pudding see PUDDING on checklist.	Tablespoons.
Salad	What ingredients? If with dressing, what type (e.g. oil, vinegar, mayonnaise, salad cream etc.)?	Tablespoons, slices (e.g. cucumber) or weight?
Sandwiches and rolls	See checklist for BREAD, ROLL, BUN, BUTTER and MARGARINE. Remember to include fillings.	
Sauce-hot	What sort: savoury or sweet? Thick or thin? Recipe or ingredients if possible? Brand?	Tablespoons.
Sauce-cold	What sort: e.g. Tomato ketchup, brown sauce, soy sauce, salad cream? Brand?	Tablespoons.

Sausages	Pork, beef, pork & beef etc.? Large or small? How cooked (grilled, fried)? Brand?	Number
Sausage rolls	Large or small? Type of pastry? Brand?	Number
Scones	With currants, sweet or plain, cheese, wholemeal? See checklist if with BUTTER, MARGARINE, JAM etc.	Number
Snacks-in packet	What sort: e.g. Cheese straws, Twiglets, pretzels, if mini-biscuits, see BISCUITS on checklist.	Packet weight
Soft drinks	What type: squash, diluted? Fizzy drinks? Normal, low-calorie or sugar free? Brand?	Glasses (small, medium, large) or cans
Soup	What sort: canned, packet, instant, fresh and home-made? Brand?	Tablespoons, bowl or mug
Soya/Quorn	TVP, mince, burgers or tofu.	Number, tablespoons or weight.
Spreads	What type? Brand? If in sandwich see SANDWICH in checklist.	½ or ¼ teaspoons: Spread thickly, average or thinly
Sugar	White, brown or Demerara?	Heaped or level teaspoons, how many?
Sweets	What sort? E.g. Toffees, boiled sweets, lollipops, bubble/chewing gum (sugar free?) etc. Brand?	Number or packet size
Tea	Include milk & sugar!	Small, medium or large cup or mug. How much milk/sugar?
Vegetables	What types? E.g. Carrots, broccoli, peas, cabbage etc. Fresh, frozen or canned. How cooked (boiled, fried, grilled and roasted)? See checklist if with BUTTER, MARGARINE or SAUCE.	Tablespoons or weight.
Water	If bottled, does it have sugar?	Glasses (small, medium, large) or bottle size
Yoghurt, Fromage Frais	What sort: e.g. Fruit, toffee, chocolate, natural, Greek, creamy or plain? Brand?	Carton weight /size or tablespoons.

WEEK ONE (from 5:00pm until arriving at school next morning) **Date:………………………..**

TIME & MEAL	DESCRIPTON OF FOOD	HOW COOKED/PREPARED	WEIGHT BEFORE (g)	WEIGHT AFTER (g)

APPENDIX I : Food preference questionnaire

What food do you like to eat?

Food Preference Questionnaire

Name……………………………………
Player number…………………………

What foods do you like to eat?	**Yes**	**No**	**Don't know**
Cheerios			
Rice Krispies			
Cheese			
Crisps			
Apples			
Pasta			
Tomato pasta sauce			
Olive oil			

What drinks do you like?	**Yes**	**No**	**Don't know**
Semi-skimmed milk			
Whole milk			
Orange juice			
Apple juice			
Water			
Apple and blackcurrant squash			

What foods and drinks do you dislike?
Are you allergic to any foods or drinks?

APPENDIX J: Methods, materials and results of the preliminary investigation to Chapter V which examined the between-day variation in fasting satiety-related peptides and glucose in 8 to 11 year-old boys.

Methods

Study design

A within-groups study design was utilised to establish the between-day variation in fasting plasma GLP-$1_{7\text{-}36}$, glucagon, insulin and leptin and blood glucose obtained from fingertip capillary blood, in 8-11 y old boys.

The study was conducted according to 2013 Declaration of Helsinki (World Medical Association. 2013) and was approved by the University of Northumbria, Faculty of Health and Life Sciences Ethics Committee. Written informed consent was obtained from the child's parent or main carer and assent was given by the child prior to data collection.

Participants

Boys aged 8-11 y were recruited from a primary school located within the city of Newcastle upon Tyne (North East England, UK) as part of the study outlined in Chapter V and described in section 5.2.3.

Study protocol

The boys were required to attend the University laboratory on two different days, separated by 1 week. From 17:00 h on each day preceding the two visits the PA, food and fluid intake of each boy was standardized, as described in Chapter IV, section 4.2.4. On the morning of each visit, following a 12 h overnight fast, the boys attended school at 0830 h. From waking, they were requested to drink only water and with the assistance of their parent or main carer were asked to note this amount in the food diary to enable replication prior to the second visit. For logistical reasons, the boys were organised into testing groups of five to seven in number. At school (0830 h), the boys were met by two members of the research team and transported to the University for 0845 h so that they could each provide one fasted capillary blood sample. In both visits, immediately following the collection of the blood sample, each boy was provided with breakfast consisting of fruit juice, cereal and toast with butter and jam. After breakfast they were escorted back to school by two researchers.

Blood sampling

Identical sampling procedures for the collection of GLP-$1_{7\text{-}36}$, glucagon, insulin and leptin and also for blood glucose were used as in Chapter IV and are described in full, in section 5.2.8.

Blood analysis: electrochemiluminescence

Forty µL of plasma was extracted from each of the stored samples to determine the concentrations of GLP-$1_{7\text{-}36}$, glucagon, leptin and insulin simultaneously, using a human hormone multiplexed sandwich immunoassay (Sector Imager 2400, MesoScale Discovery, Rockville, MD. USA). Both fingertip capillary plasma samples were analysed on one assay plate which eliminated inter-assay variation. Intra-assay coefficients of variation (CV) were established by the measurement of one baseline fingertip capillary plasma sample, three times on the assay plate. For GLP-$1_{7\text{-}36}$, glucagon, leptin and insulin, these were established as 11%, 9%, 19% and 11%, respectively. See Appendix K for in-depth assay method and materials.

The blood glucose samples were quantified by the glucose oxidase method using an automated glucose analyser (BiosenC_line, EKF Diagnostics), as in Chapter V and outlined in section 5.2.9.

Statistical analysis

For all values, means $_{\pm SEM}$ were calculated for GLP-$1_{7\text{-}36}$, glucagon, leptin, insulin and glucose. The within-subject variation between samples for visits one and two was assessed by utilising a range of statistical methods as suggested by Hopkins (2000) and Atkinson and Neville (1998). Deming regression and mean difference were the principal methods of assessment. Deming regression tests for and provides a value for average systematic and proportional bias on a group level (Deming, 1964). Mean difference, provides a value for typical error (Hopkins, 2000). The Bland-Altman limits of agreement (LOA) plots (Bland & Altman, 1999) were also used to indicate the relative bias (mean difference) and random error. The typical error as a percentage coefficient of variation (CV%) was also calculated to estimate the typical error between individuals in visit one and two. All values were checked for heteroscedasticity by the examination of box plots and scatter plots. If heteroscedasticity was apparent in the data, log transformation was performed and where necessary outliers were removed.

To aid in the interpretation of the statistical analysis, clinically significant differences deemed to be meaningful were acquired for each of the peptides and glucose in advance of data collection, based on published adult research. Adult values were utilised due to the lack of reproducibility literature to date in relation to fingertip sampling, EI or subjective appetite and GLP-$1_{7\text{-}36}$, glucagon, leptin, insulin and glucose for healthy children. The values obtained for each peptide were time-averaged AUC over 1 h and subsequently time-averaged over 90 min. In doing this the values reflected the average time of active video game play established in 7-11 y old children in Chapter III (Allsop et al., 2013) and the time duration utilised for the gaming bouts in Chapter IV (Allsop et al., 2015; 2013) which will likely be employed in Chapter V. The time-averaged AUC x 90 min values established for GLP-$1_{7\text{-}36}$, glucagon, leptin, insulin and glucose respectively, were 2.1 pg/mL (Verdich et al., 2001), 7.4 pg/mL (Cegla et al., 2013), 222.0 pg/ml (Liu, Askari, & Dagogo-Jack, 1999), 27.7 pg/mL and 0.5 mmol/L (Nair et al., 2009).

Results

Participant characteristics

A total of 23 boys took part in the study however two were excluded from the results due to non-standardisation of food intake prior to each visit. In addition, owing to issues related to blood collection, results for GLP-$1_{7\text{-}36}$, leptin, insulin and glucose are provided for 20 boys and for glucagon, 17 boys. The mean $_{\pm SEM}$ age of the boys was 10±0.2 y, stature 1.45±0.02 m, body mass 37.9±1.6 kg, waist circumference 63.7±1.65 cm and BMI 18.1±0.7 kg/m^2. According to UK age and sex-specific BMI centiles (Cole et al., 1995), the majority of boys were classified as having a healthy body mass (76.2%), 9.5% were classified as overweight and 14.3% as obese. The mean maturity offset was -0.2±0.2 y, indicating the boys were an average of 2.4 months from reaching their peak height velocity. All boys were identified as being unrestrained eaters according to the Dutch Eating Behaviour Questionnaire (van Strien & Oosterveld, 2008) with a mean $_{\pm SEM}$ dietary restraint score of 1.81±0.13, categorized as being average for boys of this age (1.53±0.06).

Agreement between GLP-1$_{7\text{-}36}$, glucagon, leptin, insulin and glucose

Deming regression

In relation to agreement between visits one and two, Deming regression analysis revealed no evidence of systematic [intercept (95% confidence interval (CI)] or proportional bias [slope (95% CI)] in the fasted plasma concentrations of any of the peptides or glucose. For GLP-1$_{7\text{-}36}$, the intercept (95% CI) was -0.1 (-2.2 to 2.1) and slope (95% CI) was 0.9 (0.5 to 1.4) (Figure 1a), glucagon intercept (95% CI) was 1.4 (-45.9 to 48.8)] and slope (95%) was 1.1 (0.7 to 1.5) (Figure 1b), leptin intercept (95% CI) was -2549 (-7260 to 2162) and slope (95% CI) was 1.5 (0.9 to 1.9) (Figure 1c), insulin intercept (95% CI) was -204.6 (-23.1 to 315.5) and slope (95% CI) was 0.8 (0.3 to 1.1) (Figure 1d) and for glucose the intercept (95%) was 2.1 (-3.6 to 7.8) and slope (95% CI) was 0.6 (-0.6 to 1.8) (Figure 1e).

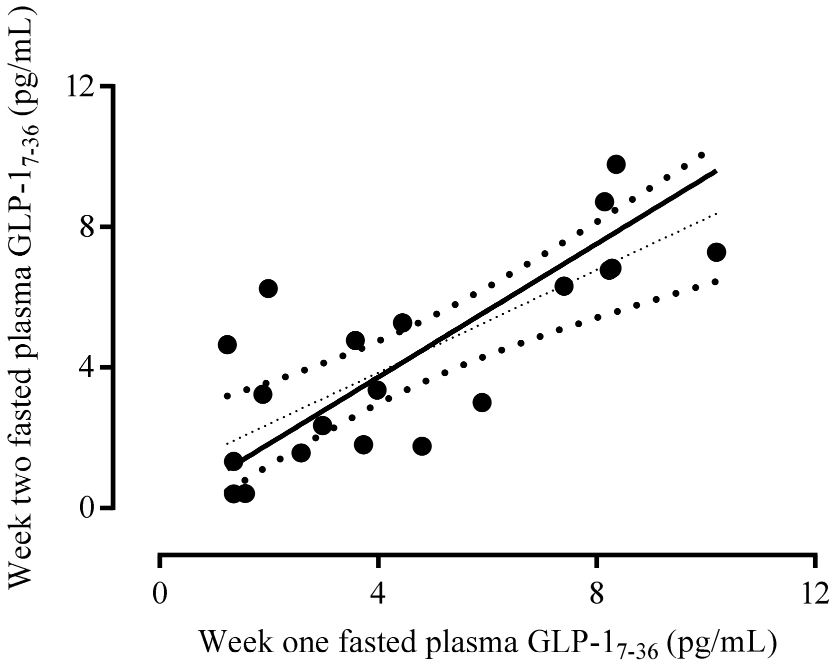


Figure 1a. Deming regression scatter-plot of fasted plasma GLP-1$_{7-36}$ for visit one versus visit two blood samples taken at same time, 1 week apart. The solid black line indicates the line of equality. The dashed line represents the regression line with the corresponding 95% CI falling in between the black dotted lines and individual data points denote the means visit one versus visit two of fasted plasma GLP-1$_{7-36}$.

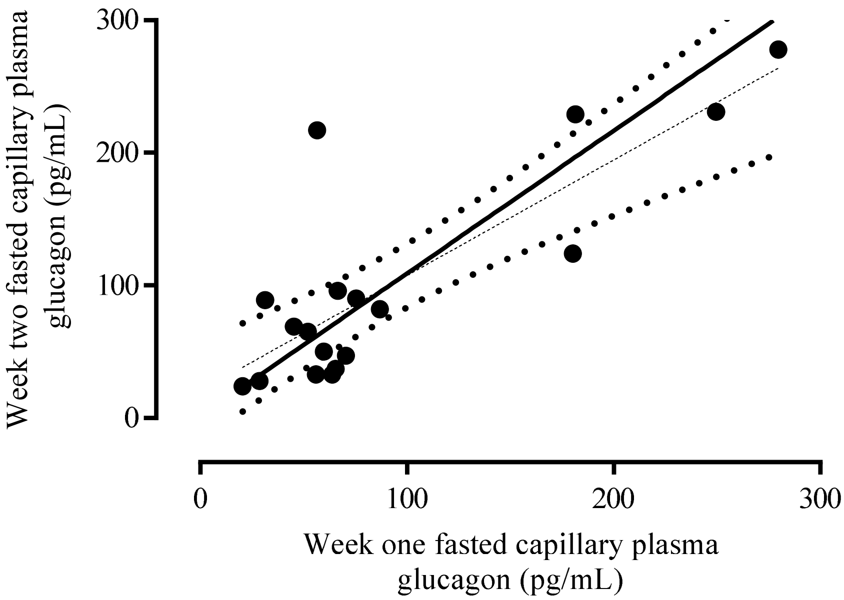


Figure 1b. Deming regression scatter-plot of fasted plasma glucagon for visit one versus visit two blood samples taken at same time, 1 week apart. The solid black line indicates the line of equality. The dashed line represents the regression line with the corresponding 95% CI falling in between the black dotted lines and individual data points denote the means visit one versus visit two of fasted plasma glucagon.

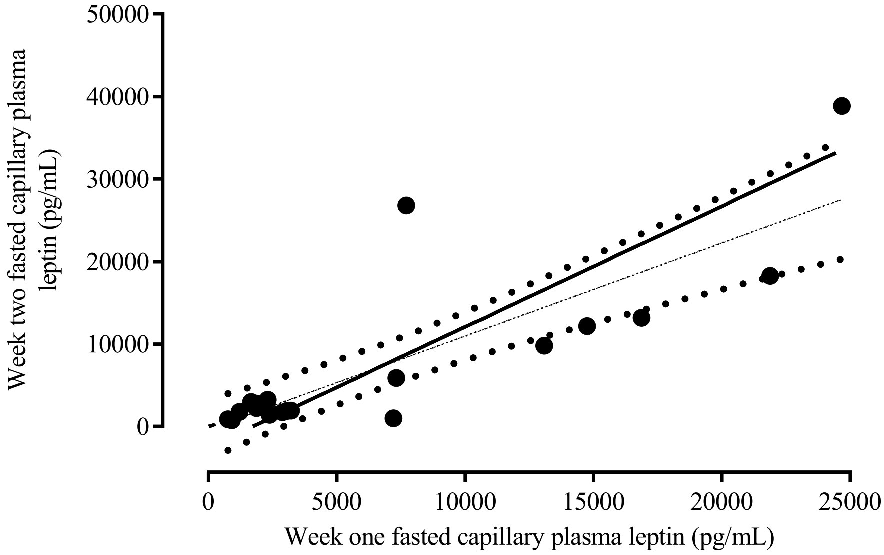


Figure 1c. Deming regression scatter-plot of fasted plasma leptin for visit one versus visit two blood samples taken at same time, 1 week apart. The solid black line indicates the line of equality. The dashed line represents the regression line with the corresponding 95% CI falling in between the black dotted lines and individual data points denote the means visit one versus visit two of fasted plasma leptin.

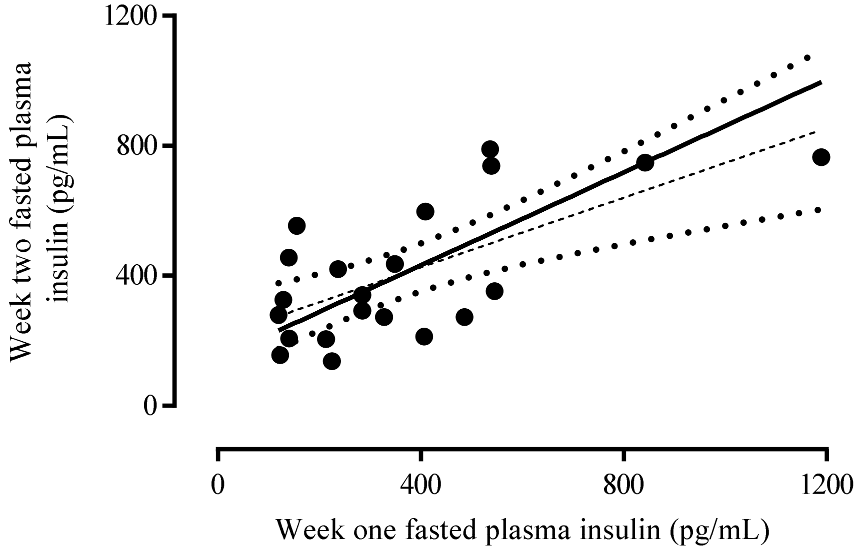


Figure 1d. Deming regression scatter-plot of fasted plasma insulin for visit one versus visit two blood samples taken at same time, 1 week apart. The solid black line indicates the line of equality. The dashed line represents the regression line with the corresponding 95% CI falling in between the black dotted lines and individual data points denote the means visit one versus visit two of fasted plasma insulin.

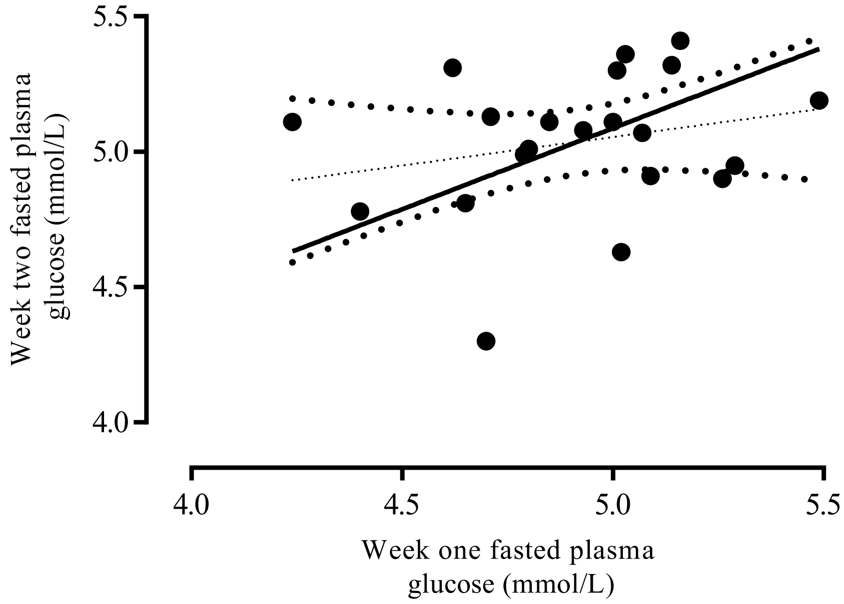


Figure 1e. Deming regression scatter-plot of fasted blood glucose for visit one versus visit two blood samples taken at same time, 1 week apart. The solid black line indicates the line of equality. The dashed line represents the regression line with the corresponding 95% CI falling in between the black dotted lines and individual data points denote the means visit one versus visit two of fasted blood glucose.

Bland-Altman limits of agreement (LOA) means, mean difference and typical error (CV%)

The mean $_{\pm SEM}$ of all peptides and glucose for visits one and two and the mean differences are displayed in Table 5.1. Typical error for each peptide expressed as a percentage coefficient of variation (CV%) are displayed in Table 1. Between visits one and two, the CV% for plasma GLP-$1_{7\text{-}36}$, glucagon, leptin and insulin were poor, although strong for plasma glucose. Bland-Altman LOA enabled the calculation between visits of relative bias (mean difference) ± random error (1.96 standard deviations (SD) of the difference). As such LOA for GLP-$1_{7\text{-}36}$ were -0.5±3.3 pg/mL (Figure 2a); glucagon 8.5±93.1 pg/mL (Figure 2b); leptin 538.3±11209.2 pg/mL (Figure 2c); insulin 41.8±390.8 pg/mL (Figure 2d) and glucose 0.1±0.7 pg/mL (Figure 2e). The LOA showed good agreement between visits one and two, for plasma GLP-$1_{7\text{-}36}$ although there was large random error. Limits of agreement for plasma glucagon, leptin and insulin exceeded the aforementioned predetermined clinical values and showed large random error. For glucose, LOA were good between visits and random error was low.

Table 1. Means $_{\pm SEM}$, mean differences $_{\pm SEM}$ and CV% between visit one and visit two for plasma GLP-$1_{7\text{-}36}$, glucagon, leptin, insulin and glucose.

	GLP-$1_{7\text{-}36}$ (pg/mL)	Glucagon (pg/mL)	Leptin (pg/mL)	Insulin (pg/mL)	Glucose (mmol/L)
Visit one					
Mean $_{\pm SEM}$	4.7±0.6	92.8±18.0	6679.8±1582.9	365.8±58.0	4.9±0.1
Visit two					
Mean $_{\pm SEM}$	4.6±0.7	101.5±19.1	7218.4±2169.4	407.9±46.2	5.0±0.1
Mean difference $_{\pm SEM}$	0.1±0.1	8.7±1.1	746.9±586.5	42.1±11.8	0.1±0.0
CV%	68.8	45.0	57.0	48.7	5.3

$_{SEM}$ standard error mean; CV% percentage coefficient of variation.

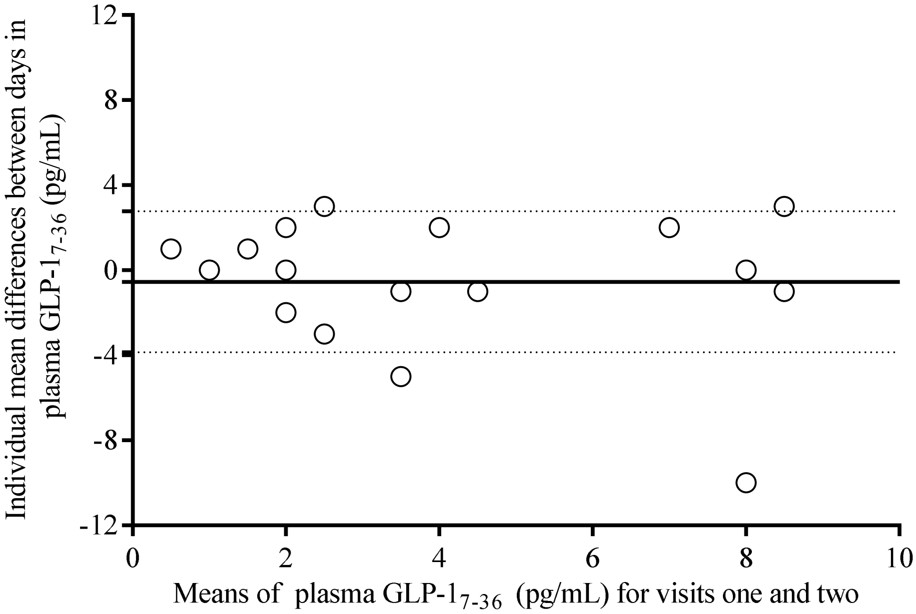


Figure 2a. Individual mean differences in plasma GLP-$_{17\text{-}36}$ plotted against the means of plasma GLP-1$_{7\text{-}36}$ between days. The solid black line represents the mean difference (bias) and the dotted lines indicate LOA (the mean difference ±1.96 SD of the mean difference).

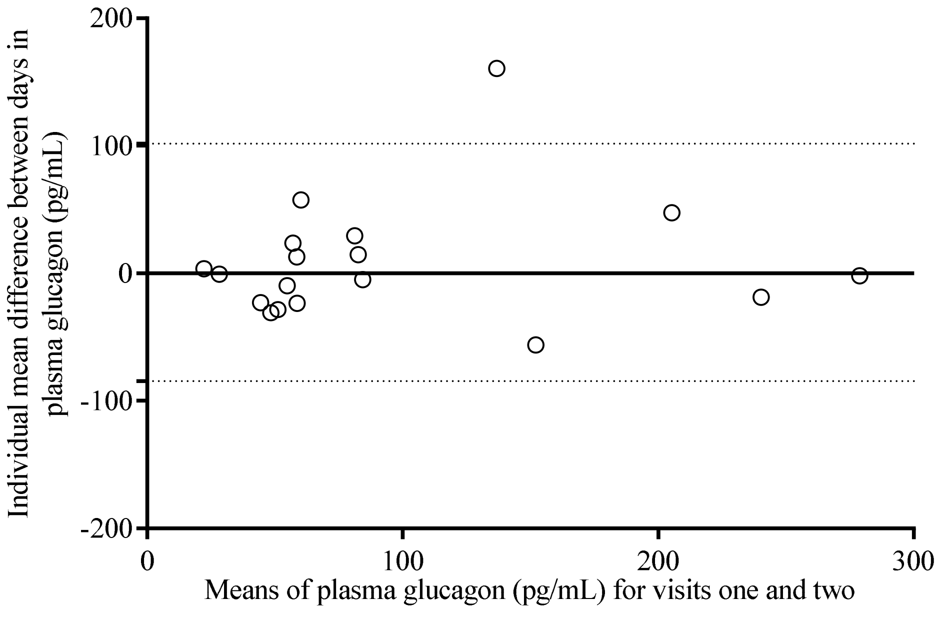


Figure 2b. Individual mean differences in plasma glucagon plotted against the means of plasma glucagon between days. The solid black line represents the mean difference (bias) and the dotted lines indicate LOA (the mean difference ±1.96 SD of the mean difference).

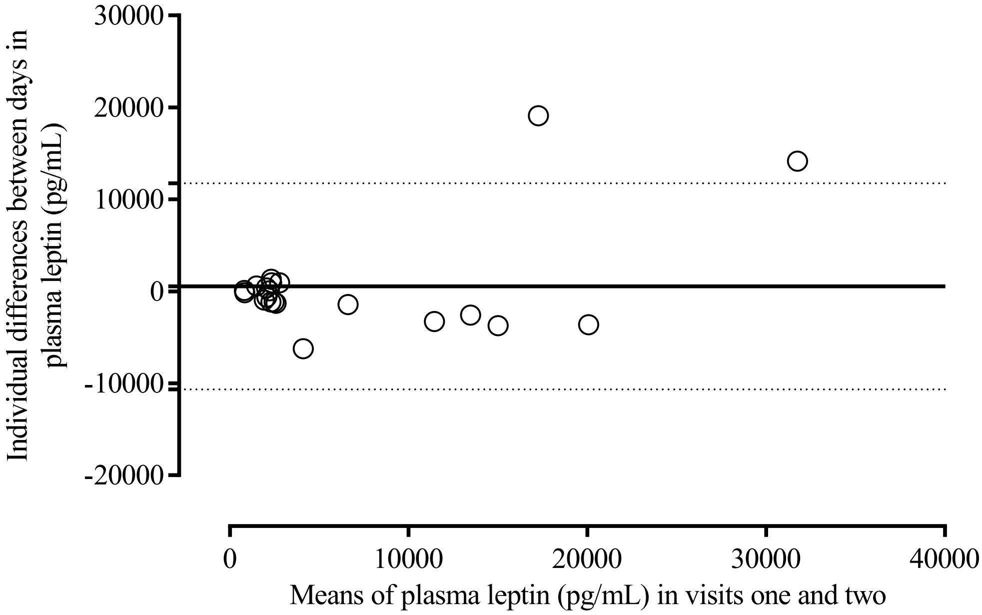


Figure 2c. Individual mean differences in plasma leptin plotted against the means of plasma leptin between days. The solid black line represents the mean difference (bias) and the dotted lines indicate LOA (the mean difference ±1.96 SD of the mean difference).

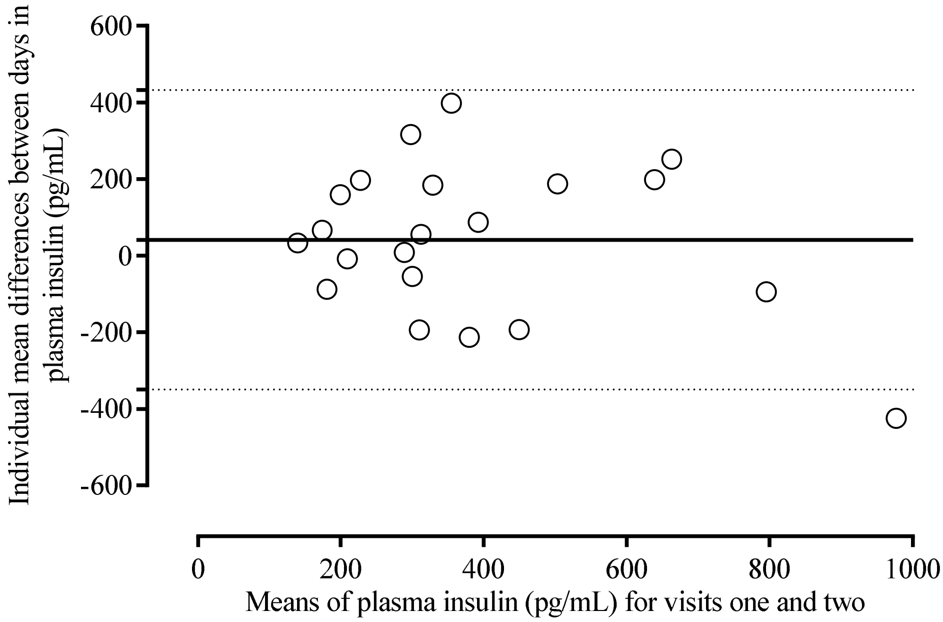


Figure 2d. Individual mean differences in plasma insulin plotted against the means of plasma insulin between days. The solid black line represents the mean difference (bias) and the dotted lines indicate LOA (the mean difference ±1.96 SD of the mean difference).

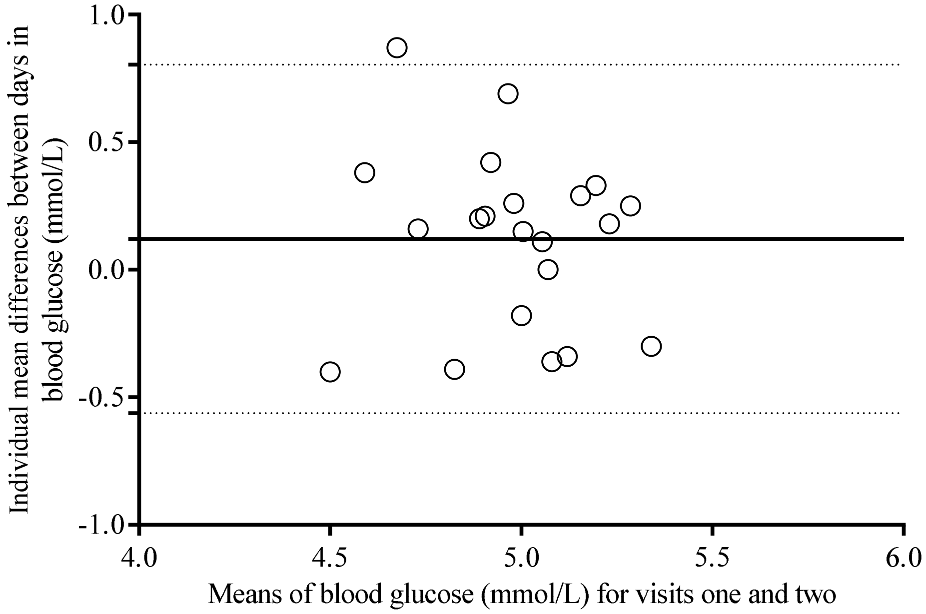


Figure 2e. Individual mean differences in blood glucose plotted against the means of blood glucose between days. The solid black line represents the mean difference (bias) and the dotted lines indicate LOA (the mean difference ±1.96 SD of the mean difference).

IV Required Materials and Equipment - not supplied

required materials and equipment – not supplied

- Deionized water for diluting concentrated buffers
- 50 mL tubes for reagent preparation
- 15 mL tubes for reagent preparation
- Microcentrifuge tubes for preparing serial dilutions
- Phosphate buffered saline plus 0.05% Tween-20 (PBS-T) for plate washing
- Appropriate liquid handling equipment for desired throughput, capable of dispensing 10 to 150 µL into a 96-well microtiter plate
- Plate washing equipment: automated plate washer or multichannel pipette
- Adhesive plate seals
- Microtiter plate shaker

V Safety

safety

Safe laboratory practices and personal protective equipment such as gloves, safety glasses, and lab coats should be used at all times during the handling of all kit components. All hazardous samples should be handled and disposed of properly, in accordance with local, state, and federal guidelines.

VI Reagent Preparation

reagent preparation

Bring all plates and diluents to room temperature.
Blockers D-B, D-R and E can tolerate up to 5 freeze-thaw cycles. Alternatively, an aliquot of each blocker can be stored at 2-8°C for up to 1 month.

> **Important:** Upon first thaw, separate Diluent 6 and Diluent 12 into aliquots appropriate to the size of your assay needs. These diluents can go through up to three freeze-thaw cycles without significantly affecting the performance of the assay.

Prepare Blocker A Solution

Follow instructions included with the Blocker A Kit.

Prepare Metabolic Assay Working Solution

In a 15 mL tube combine (per plate):

- ❑ 35 µL of Aprotinin
- ❑ 70 µL of Blocker E
- ❑ 6895 µL of Diluent 6

> **Important:** Aprotinin should be added prior to use. The Metabolic Assay Working Solution should be kept on ice. Do not freeze the Metabolic Assay Working Solution for later use.

Prepare Calibrator and Control Solutions

The stock Calibrator vials are supplied at 1 µg/mL for GLP-1 (7-36)amide and Glucagon, at 5 µg/mL for Insulin and at 10 µg/mL for Leptin. For the assay, an 8-point standard curve is recommended with 4-fold serial dilution steps and a zero Calibrator.
The table below shows the concentrations of the 8-point standard curve:

Standard	GLP-1 (7-36)amide conc. (pg/mL)	Insulin conc. (pg/mL)	Glucagon conc. (pg/mL)	Leptin conc. (pg/mL)	Dilution Factor
Stock Cal. Vial	1000000	5000000	1000000	10000000	
STD-01	10000	50000	10000	100000	100
STD-02	2500	12500	2500	25000	4
STD-03	625	3125	625	6250	4
STD-04	156	781	156	1563	4
STD-05	39	195	39	391	4
STD-06	9.8	49	9.8	98	4
STD-07	2.4	12.2	2.4	24	4
STD-08	0	0	0	0	n/a

To prepare this 8-point standard curve:

1) Prepare the highest Calibrator by adding 10 µL of 1 µg/mL GLP-1 (7-36)amide, 10 µL of 5 µg/mL Insulin, 10 µL of 1 µg/mL Glucagon and 10 µL of 10 µg/mL Leptin to 960 µL of Metabolic Assay Working Solution.
2) Prepare the next Calibrator by transferring 50 µL of the diluted Calibrator to 150 µL of Metabolic Assay Working Solution. Repeat 4-fold serial dilutions 5 additional times to generate 7 Calibrators.
3) Reserve 150 µL of Metabolic Assay Working Solution to be used as zero calibrator.
4) Diluted Calibrators should be kept on ice prior to addition to the plate.

Preparation of Serum and Plasma Samples

1) The assay format requires 40 µL of sample per well. An adequate volume of each sample should be prepared depending upon desired number of replicates.
2) There are numerous proteases in serum and plasma that may cause degradation of GLP-1 and Glucagon. Blood samples should be drawn into tubes containing 500 KIU Aprotinin per mL of whole blood. Alternately, Aprotinin should be added immediately following blood draw. Invert the blood tube several times to mix the sample.
    a. To obtain serum, tubes containing Aprotinin should be allowed to clot for 30' on a rocker. Spin the tubes for 10 minutes at 1000 x g (4°C) and aliquot serum into separate tubes and store at -80°C until use. Avoid repeated freeze-thaw (> 2) of these aliquots.
    b. Plasma samples should be obtained in vacutainer or syringe containing Na_2EDTA (1.25 mg/mL) and 500 KIU Aprotinin per mL of whole blood. Tubes should be spun for 10 minutes at 1000 x g (4°C) and then plasma immediately aliquotted into separate tubes and stored at -80°C until use. Avoid repeated freeze-thaw (> 2) of these aliquots.
3) To preserve the integrity of Active GLP-1, a DPP IV inhibitor should be added to the sample tube prior to collection.
4) Keep isolated or thawed serum/plasma samples on ice or at 4°C prior to subsequent processing or until use in the assay.
5) Samples with hemolysis or significant lipemia may hinder accurate assay measurements.

Prepare Detection Antibody Solution

The Detection Antibodies are provided as a 100X stock solution. The working Detection Antibody Solution should contain 1X as the final concentration of each antibody.
In a 15 mL tube combine (per plate):

- ❑ 90 µL of 10% Blocker D-B
- ❑ 90 µL of 10% Blocker D-R
- ❑ 30 µL of 100X SULFO-TAG Anti-GLP-1 (7-36)amide Antibody
- ❑ 30 µL of 100X SULFO-TAG Anti-GLP-1 (7-37) Antibody
- ❑ 30 µL of 100X SULFO-TAG Anti-hInsulin Antibody
- ❑ 30 µL of 100X SULFO-TAG Anti-Glucagon Antibody
- ❑ 30 µL of 100X SULFO-TAG Anti-hLeptin Antibody
- ❑ 2670 µL of Diluent 12

Prepare Read Buffer

The Read Buffer should be diluted in deionized water to make a final concentration of 1X Read Buffer T. Add 5 mL of stock Read Buffer T (4X) to 15 mL of deionized water for each plate.

Prepare MSD Plate

This plate has been pre-coated with antibodies for the analytes shown in Figure 1. The plate can be used as delivered; no additional preparation (e.g., pre-wetting) is required. The plate has also been exposed to a proprietary stabilizing treatment to ensure the integrity and stability of the immobilized antibodies.

VII Assay Protocol

assay protocol

1. **Addition of Blocker A Solution:** Dispense 150 µL of Blocker A Solution into each well. Seal the plate with an adhesive plate seal and incubate for 1 hour with vigorous shaking (300–1000 rpm) at room temperature.
2. **Wash and Addition of Sample or Calibrator:** Wash the plate 3X with PBS-T. First, dispense 20 µL of Metabolic Assay Working Solution into each well of the MSD plate. Then, immediately add 40 µL of sample or Calibrator into the appropriate wells of the MSD plate. Seal the plate with an adhesive plate seal and incubate for 2 hours with vigorous shaking (300–1000 rpm) at room temperature.
3. **Wash and Addition of the Detection Antibody Solution:** Wash the plate 3X with PBS-T. Dispense 25 µL of the 1X Detection Antibody Solution into each well of the MSD plate. Seal the plate and incubate for 1 hour with vigorous shaking (300–1000 rpm) at room temperature.
4. **Wash and Read:** Wash the plate 3X with PBS-T. Add 150 µL of 1X Read Buffer T to each well of the MSD plate. Analyze the plate on the SECTOR Imager. Plates may be read immediately after the addition of Read Buffer.

Notes

Shaking a 96-well MSD MULTI-SPOT plate typically accelerates capture at the working electrode.

Bubbles in the fluid will interfere with reliable reading of MULTI-SPOT plate. Use reverse pipetting techniques to insure bubbles are not created when dispensing the Read Buffer.

VIII Analysis of Results

analysis of results

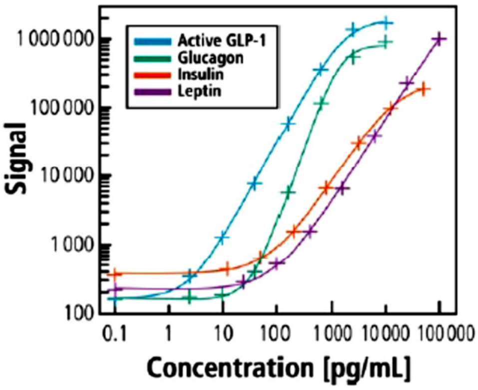
The calibrators should be run in duplicate to generate a standard curve. The standard curve is modeled using least squares fitting algorithms so that signals from samples with known levels of the analyte of interest can be used to calculate the concentration of analyte in the sample. The assays have a wide dynamic range (3–4 logs) which allows accurate quantitation in many samples without the need for dilution. The MSD Discovery Workbench® analysis software utilizes a 4-parameter logistic model (or sigmoidal dose-response) and includes a $1/Y^2$ weighting function. The weighting functionality is important because it provides a better fit of data over a wide dynamic range, particularly at the low end of the standard curve.

IX Typical Standard Curve

typical standard curve

The MSD Human Active GLP-1, Insulin, Glucagon, Leptin Assay is designed for use with human serum and plasma samples.

The following standard curves are examples of the dynamic ranges of the assay. The actual signals may vary. A standard curve should be run for each set of samples and on each plate for the best quantitation of unknown samples.



Active GLP-1		
Conc. (pg/mL)	Average Counts	%CV
0	162	10
2.4	345	1
9.8	1262	2
39	7806	1
156	57503	1
625	356515	1
2500	1345265	1
10000	1698466	1

Insulin		
Conc. (pg/mL)	Average Counts	%CV
0	362	3
12.2	437	7
49	644	4
195	1563	1
781	6666	5
3125	29947	4
12500	96447	1
50000	183660	7

Glucagon		
Conc. (pg/mL)	Average Counts	%CV
0	156	5
2.4	168	1
9.8	186	3
39	400	1
156	5724	4
625	113210	6
2500	547138	3
10000	899803	2

Leptin		
Conc. (pg/mL)	Average Counts	%CV
0	211	8
24	285	2
98	525	1
391	1572	0
1563	6585	5
6250	38369	3
25000	225767	1
100000	988180	4