An assessment of the impact of South Asian ethnicity on body composition, dimensions and proportions in children and adolescents, and their potential for risk of metabolic syndrome

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Statement of Originality

I Mahjabeen Shah, confirm that the work presented in this thesis is my own. Where information has been derived from other sources, I confirm that this has been indicated in the thesis.

Signature……

Date……..

Abstract

South Asians (SA) are a high-risk group for cardiometabolic disease, which is partly attributed, to their 'thin-fat' body composition (BC) phenotype. Generally, SAs have a higher % fat mass (FM) and less skeletal muscle mass (SMM), together with a more abdominal distribution of body fat, compared with white Europeans (WE) at equivalent body mass index (BMI) values. SAs also tend to have a shorter adult stature. Effective paediatric monitoring and clinical management requires improved tools for assessing body fatness and other components of BC. This is partly due to BMI being regarded as an inadequate indicator of adiposity and SMM, particularly for SAs, thus other field-based measures of BC have been investigated. In addition to BMI, several UK BC references for WE children and adolescents are available, including WC, %FM, and SMM; however, there are no similar references available for their SA counterparts.

This thesis is comprised of four key studies in which ethnic variations in BC (%FM and appendicular SMM (SMMa)), WC, and leg length (LL) in particular relative LL (RLL) between SA and WE children and adolescents (aged 5-18y) were investigated. The core aim was to develop age- and sex-specific percentile references for %FM, SMMa, and WC for the SA ethnicity. In study one, the BC418 bioelectrical impedance analyser (BIA) was validated against DXA for field-based BC assessment, in a sample of SA children (n= 53; 5-21y) to develop an ethnic-specific prediction equation for FM, FFM and SMMa determination, as prior studies have found BIA underestimates %FM in SAs. This equation was found to be only valid for children ≥9y, which was attributed to the opportunistic nature of recruitment. It was concluded that no single equation was valid across the whole child and adolescent population. This equation needs to be tested in an independent group to confirm its accuracy and functionality, prior to wider application. Due to discrepancies in DXA weight and scale weight, it was not possible to develop a prediction equation for SMMa, although the existing BIA output was considered acceptable due to the very small between-method relative differences.

In study two the new BIA prediction equation was applied to a large SA dataset of children (n =1,624) from low-income communities. Compared to UK90 (SDS) reference data, both SA girls and boys (9-14y) were significantly shorter, lighter, with a lower mean BMI and WC compared with their WE counterparts, with no significant differences in %FM. %FM and SMMa reference curves were constructed and comparisons were made between published WE (from affluent areas, WE_1) reference curves, together with a low-income cohort (WE_2). Comparisons in %FM at the 50th centile, between the SA and WE cohorts revealed that SAs had greater %FM overall, and this difference increased after application of the new equation. Similarly, across all age ranges SAs had significantly less relative SMMa than their WE counterparts.

The third study generated SA sex- and ethnic-specific WC centile curves. SDS comparisons with the WE_2 cohort revealed SAs had a significantly lower mean WC than their WE_2 counterparts. It was concluded that, as WC acts as a proxy for visceral fat, ethnic-specific cut-offs similar to those adopted for adults in India should be considered for children. The final study on LL revealed that SAs had a longer RLL than WE children, although as RLL data for WEs was derived, this would require further verification. The outcomes from these studies provide the evidence base and assessment tools to support the use of ethnic-specific references for children and youths in the UK from a SA background. The findings in this thesis demonstrate that overweight and obesity vary across different ethnic groups and this variation needs to be considered in the context of the clinical referral for individual children as well as for population surveillance. These are the first set of reference percentile charts for BC, proportions and dimensions in the UK SA paediatric population. Our findings support the use of these ethnic-specific references that go beyond BMI as an indicator of obesity-related metabolic health risk.

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Abbreviations

2-C	2-compartment
3-C	3-compartment
4-C	4-compartment
ADP	Air displacement plethysmography
ASD	All-schools data
BC	Body composition
BCA	Body composition assessment
BF	Body fat
BMI	Body mass index
CDC	Centers for Disease Control and Prevention
CVD	Cardiovascular disease
CT	Computed tomography
DH	Department of Health
DXA	Dual-energy x-ray absorptiometry
ECW	Extracellular water
FFM	Fat-free mass
FM	Fat mass
HOMA	Homeostasis model assessment
HSE	Health Survey for England
Ht	Height
ICW	Intracellular water
IDF	International Diabetes Federation
IOTF	International Obesity Taskforce
LL	Leg length
LM	Lean mass
LTM	Lean tissue mass
MFR	Muscle-to-fat ratio
MRI	Magnetic resonance imaging
NAO	National Audit Office
NCHS	National Center for Health Statistics
NHANES	National Health and Nutrition Examination Survey
NHS	National Health Service
NICE	National Institute for Health and Care Excellence (formerly National Institute for Health and Clinical Excellence)
NOO	National Obesity Observatory
RLL	Relative leg length
SA	South Asian
SDS	Standard deviation score
SES	Socioeconomic status
SMM	Skeletal muscle mass
SMM_a	Skeletal muscle mass appendicular
SMM_aI	Skeletal muscle mass appendicular index
STM	Soft tissue mass
T2DM	Type 2 diabetes mellitus
UNICEF	United Nations International Children's Emergency Fund
WC	Waist circumference
WE	White European
WHO	World Health Organisation
Wt	Weight

Chapter 1 Introduction

The global obesity epidemic is regarded as a major public health concern worldwide due to its associated health risks with type 2 diabetes mellitus (T2DM) and cardiovascular disease (CVD), as well as having psychosocial consequences (World Health Organisation (WHO), 2011). South Asians (SAs; from the Indian subcontinent) have been identified as having the highest risk factors for developing the diseases associated with obesity compared to other ethnic groups (Bhardwaj et al., 2008; Lear et al., 2009). The economic burden on society arising from what is largely a preventable disease is considered unsustainable (Yach et al., 2006). Hence, tackling the rise in obesity, particularly childhood obesity, which has been shown to track into adulthood (De Onis & Lobstein, 2010), is a major government priority, together with tackling health inequalities among minority ethnic groups (Department of Health (DH), 2008). This introductory chapter provides some background information that underpins the aims of this thesis and the research conducted, with a detailed literature review provided in Chapter 2.

For adult populations, Body Mass Index (BMI) defined as weight (Wt; kg)/height (Ht; m^2), is the main classification method used for determining overweight and obesity at specific cut-offs, with waist circumference (WC) used as an additional measure. Within child and adolescent populations, due to growth and development changes, sex-specific BMI centile charts based on Z-scores or standard deviation scores (SDS) are used, with the 50^{th} (or SDS 0.00) centile representing the median BMI for the population, and the 85^{th} and 95^{th} centiles used to define children that are overweight or obese respectively. In the UK, the child and adolescent reference charts are based on population data from 1990, which only included white Europeans (WEs; Cole et al., 1995). There is much research evidence that SAs compared to WEs and other ethnic groups have a much higher risk of developing T2DM and CVD at lower levels of obesity, based on the current classification system of BMI (Bhardwaj et al., 2008; Nightingale et al., 2011). The increased risk to SAs has led to much research to understand the aetiology of this increased risk. Much of this research has focussed on body composition assessment (BCA), which has identified SAs as having proportionally higher levels of fat mass (FM) and less fat-free mass

(FFM) or skeletal muscle mass (SMM) at equivalent BMI levels (Ehtisham et al., 2005), with SA adults also having greater abdominal fat accumulation, in particular visceral fat (Misra et al., 2007). These ethnic variations in body composition (BC) have been attributed to genetic dimorphism (West et al., 2013; Whincup et al.,2010), thus rising levels of obesity, particularly for SAs, is considered to have more serious health consequences, which has led to the development of simple to use and affordable BCA methods, that may be used in conjunction with BMI in wide-scale epidemiological research and in public-health practice (Pietrobelli et al., 2004), with ethnic-specific references (Haroun et al., 2010; Sluyter et al., 2010).

Bioelectrical impedance analysis (BIA), in recent years has emerged as a suitable method for assessing body composition (Pietrobelli et al.,2004), which divides the body into two components: fat mass (FM) and fat-free mass (FFM), where FM (kg) + FFM (kg) = body mass (kg). In BIA a low voltage, imperceptible electric current passes through the body, which measures the body's electrical impedance, from which TBW and FFM (as TBW is the major component of FFM and assumed to be constant) is estimated, using prediction equations validated against reference methods (Kyle et al., 2004). Technological advancements in BIA have led to the development of BIA models that are considered relatively accurate, safe, cost-effective and simple to use (Ward, 2012). The development of tetrapolar instruments that measure impedance in all four limbs as well as the trunk have enabled the measurement of segmental BC, which is useful when assessing the distribution of FM and FFM throughout the body, and making ethnic– and sex-specific comparisons (Pietrobelli et al., 2004). However, due to ethnic differences in the hydration of FFM, prediction equations should be ethnic-specific (Lohman, 1986; Wells et al., 2010). The Tanita (BC-418 MA, Tokyo, Japan) system is a widely available and relatively affordable tetrapolar BIA instrument, which is considered sufficiently accurate in measuring whole body and segmental FM and FFM. However, this model has only been validated using a WE population and research has shown it underestimates %FM in SAs (Sluyter et al., 2010). Thus one of the aims of this thesis was to develop a BIA prediction equation for SA children and adolescents, using the Tanita (BC-418 MA) model, validated against two reference methods.

Common reference methods used for validating BIA include Dual Energy X-ray Absorptiometry (DXA), Air Displacement Plethysmography (ADP), hydrodensitometry or underwater weighing (UWW), and hydrometry, which are explained in detail in the Literature Review (Chapter 2). The main focus of the validation methods reviewed in Chapter 2 is DXA and ADP, as these two methods were used in the BIA validation study (Chapter 4), although other methods are also briefly covered for the purposes of completion. Consideration is given to the benefits and limitations of these methods in terms of accuracy and precision, health and safety, cost effectiveness, and application in large epidemiological and clinical settings.

Following successful completion of the BIA validation study, a further aim of this thesis was to go towards filling the gaps in the research identified, by adding to the evidence of ethnic differences in body composition between SA children and adolescents and their WE counterparts within the UK, and to develop % FM (FM) and appendicular skeletal muscle mass (SMMa) percentile curves (Chapter 5) similar to the reference curves developed by McCarthy et al. (2006 & 2013 respectively) for WE children and adolescents. Low peripheral muscle mass is associated with an increased risk of T2DM and CVD (Bogin & Varela-Silva, 2010), which is likely due to SMM being the major route for insulin-dependent plasma glucose uptake, as well as being active in fat metabolism (McCarthy et al., 2006). The development of ethnic specific % FM and SMMa percentile curves offer the potential of identifying those children and adolescents that may be at increased risk of developing T2DM, who would otherwise be considered 'healthy' based on the current BMI centile cut-offs. The development of SA percentile curves for %FM and SMMa (using data from all four limbs), will provide important information on the differences in BC between these ethnic groups, and will enable health practitioners to make more informed decisions on further assessments that may be required and take any necessary preventative action.

As stated above, for adult populations, WC cut-offs are used as an additional measure in conjunction with BMI to determine the health risks associated with

overweight and obesity. However, in the UK the National Institute for Health and Care Excellence (NICE, 2013) does not recommend the use of WC as a routine measure for determining obesity in children and adolescents, but advises that it may be used to gain additional information for assessing long-term health risks, and also recommends that in the absence of ethnic-specific reference standards, the BMI UK90 charts should be interpreted with caution in ethnic minority groups. Gender- and ethnic-specific nationally representative WC percentile charts for children and adolescents have been developed both in the UK (McCarthy et al., 2001) and worldwide. The UK WC centile charts are based on WEs only, and there are currently no WC charts for SA children. Thus to address this gap in the research, a study was conducted (Chapter 7) to develop WC centiles for SA children and adolescents in the UK, with comparisons made to the WE centile charts developed by McCarthy et al. (2001).

In addition to BC as an assessment for health risks associated with obesity; body size and proportionality, in particular stature and relative leg length are also used as indicators of growth and for monitoring health (Bosy-Westphal et al., 2009). Taller-for-age children have a greater propensity to be overweight or obese. Shorter leg length relative to height, often referred to as a low leg-length-to-height-ratio (LLHR), or high sitting-height-ratio (SHR), has also been identified as a risk factor for obesity and its related diseases, with evidence that taller-for-age children may have a low LLHR (Pliakis & McCarthy, 2010). In healthy populations, the environment exerts a greater influence on body proportionality than genetics (Bogin & Varela-Silva, 2010). However, there is also considerable evidence of differences in body size and proportionality between ethnic groups worldwide (Dangour et al., 2002; Eveleth and Tanner, 1976). Due to secular increases in body stature and leg length, the UK 1990 reference standards for WE children were updated in 2002 (Dangour et al). There are currently no similar standards for SA children, thus in order to make stature and relative leg length comparisons between SA and WE children, and review any possible health implications, a study was conducted (Chapter 8) to measure stature and relative leg length, determined by measuring sitting height (SH). Due to the lack of a valid field-based tool for

measuring SH, a validation study was conducted (Chapter 6) following the modification of the Leicester Height Measure.

Research has also shown within-group differences among SAs in health risks associated with obesity, which have been related to differences in BC (Whincup et al., 2010), and body size and proportionality (Kelly et al., 1997). Thus, where possible, within-group comparisons were also made in the relevant studies. The general methods used in all the studies in this thesis together with details of the common statistical methods used, are explained in Chapter 3. The final overall conclusions and summary of research conducted, together with study limitations and recommendations for future work are detailed in Chapter 9.

Chapter 2 Literature Review

2.1 Body Composition & Related Health Risks

It is established that excess body fat, often described as overweight and obese, increases the relative risk of morbidity and mortality from many chronic non-communicable diseases; in particular type 2 diabetes mellitus (T2DM) and cardiovascular disease (CVD; McArdle et al., 2007; Dulloo et al., 2010; Nishida et al., 2010). Accumulation of excess body fat is attributed to an energy imbalance, where energy intake regularly exceeds energy expenditure (McArdle et al., 2007). However, the health risks associated with excess fat stored in adipose tissue vary according to the distribution of fat within the body, with increased health risks associated with excess fat within the abdominal or trunk area, in particular visceral fat deposition, often described as "android" or central obesity, as opposed to fat distributed within the hip and thigh regions, described as "gynoid" obesity (IDF, 2006; McArdle et al., 2007; McCarthy, 2006; WHO, 2000).

It is important to define healthy ranges for % fat mass (FM), i.e. the minimum and maximum levels of %FM required for the body to function normally, with levels outside of this range deemed to put the individual at risk for disease (Helba & Binkovitz, 2009; Heyward & Wagner, 2004). Thus it would be inappropriate to use 'average' values for this measure, particularly in a population that has an increasing proportion of overweight and obese individuals. According to Lohman et al. (1997), for men and women respectively the average %FM is 13% and 28%; with levels of FM >25% for men and >35% for women regarded as obese; with minimum healthy fat levels of 8% for men and 20% for women. It is more difficult to define healthy %FM ranges for children, as in general the consequences of excess fat in terms of the development of disease are not realised until adulthood. For females, a minimum of 17%FM has been purported as necessary for the initiation of menses (Frisch, 1987), and at least 22%FM for normal ovulatory function (Heyward & Wagner, 2005; Frisch & Revelle, 1970).

For children (10-16y), 'metabolic syndrome' is defined by the International Diabetes Federation (IDF; Alberti et al., 2007), as a cluster of risk factors for CVD and T2DM,

including abdominal obesity with a population-specific waist circumference cut-off and at least two other clinical measures (see section 2.3.2). The IDF recommend use of the adult criteria for metabolic syndrome (IDF, 2006) for children ≥16y, and for children 6 - <10y it recommends that weight (Wt) reduction be advised for those with abdominal obesity (see section 2.3.2; Zimmet et al., 2007). Metabolic syndrome leads to a 2-3 fold increase in the risk of CVD, and a 5-fold increase in the risk of developing T2DM (Alberti et al., 2005). Children under 6y were excluded from the IDF definition due to insufficient data available for that age group.

2.2 The Obesity Related Health Risks to South Asian Children and Adolescents

Obesity is a chronic non-communicable and largely preventable condition, which due to its high prevalence worldwide is now regarded as a global epidemic; over the last 25 years, child and adult obesity has dramatically escalated, having more than doubled since 1980 (WHO, 2000). In 2008, the World Health Organisation (WHO, 2011) estimated 1.5bn adults (≥20y) globally as overweight, with over 1 in 10 adults and almost 43m children (>5y) as obese.

Rising levels of childhood overweight and obesity in the UK and worldwide are a major public health concern (Bhardwaj et al., 2008; Butland et al., 2007; Saxena et al., 2004), and tackling this rise in obesity is a major government priority, with its main focus on childhood obesity (DH, 2008). Childhood overweight and obesity is strongly linked to long-term morbidity and mortality risks such as T2DM and CVD that are independent of body mass in adulthood (Murasko, 2009; NHS Information Centre, 2010; Saxena et al., 2004; Wang, 2001).

In report from the International Association for the Study of Obesity/International Obesity Task Force (IASO/IOTF, 2010) it was estimated that worldwide, up to 200m school-aged children were either overweight or obese, out of which approximately 40-50m were classified as obese, based on body mass index (BMI) measures. Within the EU, it was estimated that approximately 60% (~260m) of adults and over 20% (~12m) of school-aged children were either overweight or obese (IASO/IOTF, 2010). The 2009 Health Survey for England (HSE; Aresu et al., 2010) classified 61.3% of adults (≥16y) and 28.3% of children (31% boys vs 28% girls aged 2-15y) in England as overweight or obese (based on BMI ≥85th <95th percentile for

overweight; ≥95th percentile for obese), of which 23% of adults and 14.4% of children were classified as obese. More recent data shows that levels of overweight or obesity are continuing to rise, with results from 2014 revealing that 61.7% of adults (≥16y) and 31.2% of children (2-15y) were overweight or obese (Public Health England, 2016). In the 2004 Health Survey for England (HSE) on minority ethnic groups (Sproston & Mindell, 2006), prevalence data on overweight and obesity for South Asian (SA) children and adolescents aged 2-15y by specified SA ethnicity (i.e. Indian, Pakistani, and Bangladeshi), together with data for the general population, was provided. In that report 30% of boys and 31% of girls in the general population were either overweight or obese, compared to 26% and 31% (Indian); 39% and 25% (Pakistani); 34% and 33% (Bangladeshi) of boys and girls respectively. Pakistani boys were identified as having one of the highest proportions of overweight or obese children, with other ethnic groups not significantly different to the general population (Sproston & Mindell, 2006).

Compared to white Europeans (WE), SAs have been identified as a high-risk group for morbidity and early mortality from CVD and T2DM (Bhardwaj et al., 2008; Bhopal et al., 1999; Chaturvedi, 2003; Whincup et al., 2002), which is attributed to a tendency for central obesity and increased visceral and subcutaneous fat underpinned by a genetic predisposition to the 'metabolic syndrome' (Bhardwaj et al., 2008; Chaturvedi, 2003; Lear et al., 2009; Misra et al., 2007; Saxena et al., 2004; Whincup et al., 2002). In addition to the measures defined by the IDF (2007) for diagnosing metabolic syndrome, other studies have reported SAs to have raised levels of C-reactive protein, and higher levels of fasting insulin, which are strongly associated with insulin resistance and increased risk of T2DM and CVD (Bhardwaj et al., 2008; Misra et al., 2007; Whincup et al., 2010). Recent studies comparing body composition (BC) of SA and WE children and adolescents in the UK have revealed that SAs have greater body fat than their age and sex matched WE counterparts (Ehtisham et al., 2005; Nightingale et al., 2011), with greater central adiposity and reduced insulin sensitivity, which was directly related to the differences in adiposity (Ehtisham et al., 2005). However, not all studies comparing WE and SA children and adolescents have identified significant differences in adiposity between the two ethnicities based on BMI or waist circumference (WC) measures alone (Bhopal et

al., 1999; Wardle et al., 2006). For example, compared to the general population, the 2004 HSE (Sproston & Mindell, 2006) found only Pakistani boys had a significantly greater proportion of overweight or obese children, with no significant differences between other SA ethnic sub-groups.

T2DM is estimated to represent around 90% of all cases of diabetes (NICE, 2012). In 2012 the annual cost to the NHS associated with diabetes was approximately £8.8 billion, which amounted to just over 8% of its total annual budget (NICE, 2012). The prevalence of T2DM had more than doubled from 1996 to 2.6 million in 2009, (Diabetes UK, 2010), was approximately 3.1million in 2010, and is projected to rise to 4.6 million by 2030 (NICE, 2012). In the UK, SAs are reported to have a six-fold greater risk of developing T2DM than white Europeans (Ehtisham et al., 2000).

T2DM was until relatively recently limited to adulthood; however, in 2000 the first cases were diagnosed in overweight and obese SA and Arabic children (Ehtisham et al., 2000). Based on minimum prevalence estimates from a survey conducted in 2000 of all UK-based paediatric diabetes centres, it was reported that SA children (<16y) had a 13.7 times greater relative risk of developing T2DM than white UK children (Drake et al., 2002; Ehtisham et al., 2004). Whilst the prevalence of T2DM during childhood is still relatively uncommon (Haines et al 2007), studies have found precursors of T2DM or symptoms of metabolic syndrome in children, such as dyslipidemia and impaired fasting glucose, with a much higher prevalence of these precursors found in SA children compared to WE children, particularly children of a Bangladeshi origin, which reflect prevalence rates of T2DM in adults (Whincup et al., 2010). These findings clearly support the premise that early intervention strategies to prevent overweight and obesity in childhood, particularly among SAs are vital for the current and future health status of the population (Misra et al., 2007; Murasko, 2009; Saxena et al., 2004; IDF, 2007). In recognition of ethnic differences in health risk and outcome, tackling health inequalities among ethnic minority groups is a government priority (DH, 2001).

Within the UK, black African-Caribbean adults have a 2-fold greater risk of developing T2DM compared with WE adults; although some of the underlying

metabolic risk factors tend to be quite different to those present within SAs, in particular the blood lipid profiles (Whincup et al., 2010). However, both populations tend to have higher levels of adiposity (Whincup et al., 2010) than white Europeans, which suggests that certain risk factors for developing T2DM can vary between ethnic groups and should be considered separately.

Studies also show heterogeneity in overweight, obesity, and associated health outcomes within SA groups as well as between genders (Ehtisham et al., 2005; Lear et al., 2009; NHS Information Centre, 2006; Shaw et al., 2007; Whincup et al., 2010) which may be associated with differences in parental body fat patterning, race/ethnicity, religion and culture, diet, physical activity behaviours and socioeconomic status, among others (Bhopal et al., 1999; Magnhild et al., 2008; Murasko, 2009; Owen et al., 2009; Saxena et al., 2004). Whilst diet and physical activity have been considered as strong contributory factors to body composition within ethnic sub-groups (NHS Information Centre, 2006), the differences in body composition between ethnic groups are likely to be evident from early life; as much evidence shows that even as babies SAs have higher %FM than WE babies of a similar Wt and body size, and lower levels of lean mass (LM; Lear et al., 2009; Stanfield et al., 2012; West et al., 2013), which may be related to in-utero factors (Stanfield et al., 2012; West et al., 2013; Whincup et al., 2010).

Ethnic differences in the precursors of T2DM including anthropometric and blood sampling measures were carried out in a recent school-based Child Heart and Health Study (CHASE Study) involving 4,796 apparently healthy British children aged 9-10y from 200 schools in three major UK cities (183 London, 14 Birmingham, and 3 Leicester) with a large percentage of children with a SA (n= 1,306) or black African-Caribbean origin (Whincup et al., 2010). To ensure greater independence of height (Ht) in this study, ponderal index (Wt kg/Ht m^3) was used instead of BMI due to evidence of its unreliability when comparing ethnic groups (WHO, 2004) and FM was determined by two methods: sum of four skinfolds, and leg to arm BIA with DXA validated equations for determining FM in children, which was presented as a FM index (FMI; FM/Ht^2).

Differences in anthropometric and FM measures between SA and WE children revealed similar height, lower Wt, mean ponderal index and WC, but a higher mean sum of skinfolds and FMI in the SA group. Additionally, all blood-sampling measures presented higher risk factors for T2DM (including higher levels of fasting glucose and insulin, C-reactive protein and triglycerides). However, adjustment for adiposity including sum of skinfolds and FMI had little effect on the blood markers related to the risk for T2DM, which suggests that FM per se is not the main contributor to the risks for T2DM in children, particularly in normal Wt/BMI SA children. Other studies in SA children have also found that adiposity does not directly correlate with insulin resistance (Whincup et al., 2002). Differences in LM were not presented in this study (Whincup et al., 2010), however, considering that the SA children were on average lighter than the WE children, yet had a higher FMI, suggests that LM was likely to be lower and may be a significant contributor to the risk factors identified in the blood-sampling measures, as has been identified in previous studies (Lear et al., 2009; Rush et al., 2009; Stanfield et al., 2012).

Differences within SA groups for all measures revealed that Bangladeshi children had a more adverse health profile than Pakistani and Indian children in terms of specific measured risk factors for T2DM (Whincup et al., 2010). This study adds to the evidence that the increased risk of T2DM within SAs is present in childhood and there are within group differences, as seen in this study with Bangladeshi children at an even greater risk, which is consistent with reports of the very high prevalence of T2DM and CVD among Bangladeshi adults (Bhopal et al., 1999; Erens et al; 2001; Sproston & Mindell, 2006); although as stated previously, this may be related to ethnic sub-group differences in diet and physical activity. Evidence of such differences in diet and physical activity were reported in the HSE 2004 (Sproston & Mindell, 2006), with Bangladeshi adults consuming diets with the highest fat and lowest fibre content, together with the lowest levels of physical activity compared to Indian and Pakistani sub-groups. Interestingly, Bangladeshi adults had the lowest prevalence of obesity based on BMI >30kg/m^2, which illustrates the inadequacy of BMI as a proxy measure for obesity. Furthermore, of the three SA ethnic sub-groups, Indians had the lowest prevalence of T2DM and CVD, with the highest levels of physical activity, and lowest levels of dietary fat and low fibre consumption, which

highlights the importance of diet and physical activity in reducing the prevalence of T2DM and CVD in this high-risk population.

The findings reported above lend support to the argument that Wt for Ht indices such as BMI do not accurately reflect body composition within the SA paediatric population, and are not reliable indicators of the risk of developing T2DM, particularly within normal BMI ranges. In addition, adjustment for socioeconomic status in the study conducted by Whincup et al. (2010) had little effect on ethnic differences for the measured outcome variables, which has been observed in other studies (Lear et al., 2009).

A study conducted by Lear et al. (2009) on third generation Canadian male and female immigrants of SA, Aboriginal, Chinese, and European origin, with approximately 100 participants from each sex and ethnic group (mean age 44-50.7y), revealed that compared to the other three ethnic groups, SAs had significantly less LM at any given fat mass (FM) value, with a higher FM to LM (FM: LM) ratio after adjustment for possible confounding variables (age, height, frame size, sociodemographic and lifestyle factors), FM: LM for SA men was higher than that for Chinese and European men, and for SA women remained higher than all three ethnic groups. Both SA men and women had the highest insulin and HOMA levels even after adjustment for all variables including total body FM, although in certain cases differences between SAs and Aboriginals were not significant after adjustments. However, a key finding of this study was that no differences in insulin or HOMA levels were found when adjusted for F: LM (Lear et al., 2009). Similarities in outcome between SA and Aboriginals, was attributed to a common ancestry between the two ethnic groups. This study provides evidence that LM may play a significant role in determining the level of risk associated with T2DM, given that other studies have also shown that even after adjustment for FM and fat distribution, SAs have a greater propensity for insulin resistance (Chandalia et al., 1999).

In another adult multi-ethnic BC study using DXA, compared to Polynesians and Europeans, Asian Indians at lower BMI values (24 and 26 kg/m^2 for males and females respectively) had the same %FM as the other ethnic groups with BMI values

≥30 (Rush et al., 2009). Overall Asian Indians had greater total and abdominal fat, with less LM and bone mineral than the other ethnic groups. Furthermore, in Asian Indians abdominal fat increased with increasing age without a concomitant increase in total %FM, whereas in the other ethnic groups both abdominal and total %FM increased with increasing age.

Given that LM, predominantly skeletal muscle, is a major site for insulin mediated glucose uptake (McArdle et al., 2007; Miller et al., 1984), and adipose tissue exhibits insulin resistance due to its reduced density of insulin receptors (McArdle et al., 2007), it is unremarkable that the SA phenotype of a ratio of high FM to LM lends this ethnic group to a greater propensity to develop T2DM than other ethnic groups, in particular white Europeans. Dulloo et al. (2010) provide a strong argument for the use of a fat and fat free mass index (FMI and FFMI respectively), which would provide a more useful comparison of body composition between individuals of different heights than BMI or %FM. Unlike BMI, such measures would be less likely to classify highly muscular individuals as overweight or obese, and also more importantly would also help to identify individuals and populations that may be at risk of T2DM due to a high FMI or low FFMI that fall within the normal BMI range.

Use of the FFMI and FMI has been considered in childhood, with proposed gender and age-adjusted charts that take account of differences in growth and development during childhood and between sexes (Wells, 2000). Use of this method whilst promising, requires the use of a valid, widely available and relatively affordable, field-based body composition assessment tool. In recent years this method of assessment is being increasingly evaluated (Demerath et al., 2006; Eissa et al., 2009; Nakao & Komiya, 2003).

2.3 Body Mass Index and Waist Circumference (BMI and WC)

The body mass index (BMI) is a proxy measure for determining whether an individual is underweight, normal weight, overweight, or obese, and is the most commonly adopted method (NICE, 2006; NHS Information Centre, 2011), internationally defined by the World Health Organisation (WHO, 2000) as a measure of Wt relative to Ht (kg/m^2). This classification method, whilst crude, as it does not measure adiposity directly, or the distribution of adiposity throughout the body, and

cannot differentiate between individuals of high adiposity versus high muscle mass, is widely used as it is non-invasive, involving simple and common measurements, and is considered robust in identifying obese individuals, particularly at a population level (NHS, 2011; WHO, 2000). BMI has been described as having 'high specificity' for overweight and obesity (i.e. very few individuals that are defined as overweight or obese by BMI, would be neither overweight nor obese according to more direct measures of adiposity), but 'low sensitivity' (i.e. a number of individuals that are over-fat according to more direct measures of adiposity are not identified by BMI (Pliakas & McCarthy, 2010). This latter point is of particular concern to the SA population, as research has consistently reported anthropometric differences in SA children including babies (Haroun et al., 2009; Misra et al., 2003; Rush et al., 2004; Whincup et al., 2007), and adults (Dulloo et al., 2010; Misra et al., 2007), with higher levels of percentage body fat, in particular greater central adiposity including visceral and truncal fat (Dulloo et al., 2010; Saxena et al., 2004; Viner et al., 2010) and also less LM (Lear et al., 2009) at equivalent BMIs, compared to white Europeans.

The risk of morbidity and mortality increases with increasing percentage FM and decreasing FFM relative to height (FFMI; kg/m^2), as well as the distribution of fat, with central adiposity or a raised WC, associated with an increased risk for metabolic syndrome; therefore, such measures are often recommended in addition to BMI, in clinical and health settings (Bosy-Westphal et al., 2009; Cole et al., 2000; IDF, 2006; McCarthy et al., 2003; Sun et al., 2003; WHO, 2004). Whilst WC cannot differentiate between subcutaneous and visceral adipose tissue, which would require far more advanced lab-based procedures such as CT or MRI scanning (Sniderman et al., 2007), it is considered a very simple and accurate marker for central obesity, being both a highly specific and sensitive (McCarthy, 2006).

2.3.1 Adult Cut-off Points – BMI and WC

For adults in general the BMI classifications are defined as: 18.5-24.9 for healthy Wt; 25–29.9 for overweight; 30-34.9 for obesity I; 35-39.9 as obesity II; and ≥40 as obesity III (NICE, 2006; WHO, 2000), with the overweight and obese cut-off values associated in particular with morbidity and mortality risk from type 2 diabetes and cardiovascular diseases (Dulloo et al., 2010; Whitlock et al., 2009; WHO, 2000, &

2004). The BMI cut-off points defining overweight and obesity have been validated by data based mainly on white populations from Europe and the USA (WHO, 1995). However, it is now well recognized that these cut-off points are not appropriate for all ethnic groups, particularly SAs, who as stated above exhibit risks for obesity related diseases below the WHO BMI cut-offs (Gray et al., 2011; Liu et al., 2011; Luke, 2009; Misra et al., 2009). To address this issue, in 2002, a WHO Expert Committee convened to review the scientific evidence and make appropriate recommendations (WHO, 2004).

Whilst the Expert Committee (WHO, 2004) concluded that Asians in general had a higher body fat percentage, and substantially higher risk factors for obesity related diseases even below the BMI cut-off of ≥25kg/m^2, no single overweight or obese cut-off could be recommended, as the risks varied between Asian groups, which would further make comparisons between population groups difficult. The Committee thus recommended that the international BMI cut-off points should continue to be used, alongside additional trigger BMI cut-points for public health action for Asian populations, emphasising that BMI should be regarded as a continuum, with cut-offs as a guide for public health and clinical use. The trigger points for Asians were: 18.5 to <23kg/m^2 increasing but acceptable risk; 23 to <27.5kg/m^2 increased risk; and ≥27kg/m^2 as high risk. The Committee also recommended that WC measures with appropriate cut-off points be used in addition to BMI, in populations with a propensity to central obesity and related risks of metabolic syndrome.

In 2008, WHO convened an expert consultation on Waist Circumference and Waist-Hip Ratio, where recommendations for BMI cut-offs alongside WC cut-offs were made (WHO, 2011; see Table 2.1). The WC cut-offs of >102cm for men and >88cm for women are based on data mainly from WE populations, and whilst ethnic specific cut-offs were considered by the Expert Committee, due to a lack of data in other population groups, the Committee decided against this (WHO, 2011).

Table 2.1: WHO recommendations for BMI & waist circumference cut-offs (WHO, 2011)

Table 5.1 Combined recommendations of body mass index and waist circumference cut-off points made for overweight or obesity, and association with disease risk

	Body mass index	Obesity class	Disease risk (relative to normal weight and waist circumference)	
			Men < 102 cm Women < 88 cm	Men >102 cm Women >88 cm
Underweight	<18.5			
Normal	18.5–24.9			
Overweight	25.0–29.9		Increased	High
Obesity	30.0–34.9 35.0–39.9	I II	High Very high	Very high Very high
Extreme obesity	>40.0	III	Extremely high	Extremely high

Source: NHLBI Obesity Education Initiative (2000)

WHO = World Health Organisation; BMI = body mass index

In 2009, the Indian Health Ministry formally adopted new obesity guidelines, following release of a 'Consensus Statement' by the Association of Physicians in India (Misra et al., 2009) with BMI and waist circumference cut-offs based on morbidity risks associated with obesity, for the Indian population. The BMI cut-offs were: 18-22.9 for normal weight; 23-24.9 for overweight; and ≥25 for obese. Two waist circumference cut-offs were proposed at 'Action level 1': >78cm and > 72cm for men and women respectively, where any individual above these cut-offs should be advised to avoid gaining weight and maintain physical activity, although it was stated that further research was required on these cut-offs; and 'Action level 2': >90cm for men and >80cm for women should be advised to seek medical help for investigating and managing morbidity risk associated with obesity.

In the UK, the National Institute for Health and Care Excellence (NICE, 2006, 2013) published its latest BMI guidance on obesity, which is as stated above, however, NICE recognized that these cut-off points may be less accurate for certain population groups such as Asians, and recommended use of 'caution and clinical judgment' by healthcare professionals. NICE (2006, 2013), also recommended use of waist circumference alongside BMI, when assessing health risks associated with overweight and obesity (Table 2.2).

Table 2.2: NICE BMI & waist circumference cut-offs

BMI classification	Waist circumference		
	Low	High	Very high
Overweight	No increased risk	Increased risk	High risk
Obesity I	Increased risk	High risk	Very high risk
For men, waist circumference of less than 94 cm is low, 94–102 cm is high and more than102 cm is very high For women, waist circumference of less than 80 cm is low, 80–88 cm is high and more than 88 cm is very high			

Obesity: the prevention, identification, assessment and management of overweight and obesity in adults and children. Source: Copied from National Institute of Health Care Excellence (NICE;2006). BMI = body mass index

In 2006, the International Diabetes Federation (IDF) issued its guidelines for WC cut-offs based on risks associated with central obesity and the metabolic syndrome (Table 2.3). In these guidelines, the WC cut-offs for SAs were equivalent to those adopted by India at 'Action level' 2 (Misra et al., 2009), and whilst the IDF and NICE (2006) WC cut-offs for women were identical regardless of ethnicity, the WC cut-offs for men differed, with NICE (2006) stating that a WC <94cm represented a low risk, whilst the IDF (2006) and Indian (Misra et al., 2009) guidelines defined SA men with a WC of ≥ 90cm as having central obesity. The WHO (2011), on the other hand, advised that a WC of <102cm and <88cm in men and women respectively regardless of ethnicity, should only be deemed an increased, high, very, or extremely high risk, in combination with the respective BMI categories for overweight to extreme obesity. Whilst the different cut-offs for BMI and WC are somewhat confusing, it would seem prudent for health practitioners in the UK, to err on the side of caution, and assess obesity-related health risks for SAs at the lower BMI and WC cut-offs.

Table 2.3: IDF Waist Circumference Cut-offs

Table 2: Ethnic specific values for waist circumference

Country/Ethnic group		Waist circumference
Europids* In the USA, the ATP III values (102 cm male; 88 cm female) are likely to continue to be used for clinical purposes	Male	≥ 94 cm
	Female	≥ 80 cm
South Asians Based on a Chinese, Malay and Asian-Indian population	Male	≥ 90 cm
	Female	≥ 80 cm
Chinese	Male	≥ 90 cm
	Female	≥ 80 cm
Japanese**	Male	≥ 90 cm
	Female	≥ 80 cm
Ethnic South and Central Americans	Use South Asian recommendations until more specific data are available	
Sub-Saharan Africans	Use European data until more specific data are available	
Eastern Mediterranean and Middle East (Arab) populations	Use European data until more specific data are available	

The International Diabetes Federation (IDF) consensus worldwide definition of the metabolic syndrome. Source copied from: IDF (2006)

2.3.2 Child and Adolescent Cut offs – BMI and WC

Among children and adolescents, the classification of overweight and obesity and the associated risks of morbidity with specific BMI values are more complex due to large variations in growth patterns during childhood as well as between genders (Cole et al., 1995; NHS Information Centre, 2011). This has led to the development, by a number of countries, of sex-specific BMI percentile curves, with BMI cut-offs for overweight and obesity based on nationally representative data (Cole et al., 1995; WHO, 2000). However, many of these have been criticised due to the data used not being sufficiently contemporary and thus not representative of the current population, or due to the use of a restricted age range (WHO, 2000). The shape of the BMI centile curves indicates that BMI rises rapidly during the first year of life, declines until the age of approximately 5 years, and then continues to rise into adulthood (Cole et al., 1995; Pliakas & McCarthy, 2010). This rise in BMI from age 5 years has been termed the 'adiposity rebound' (Rolland-Cachera et al., 1984), which

is considered to be the age when adiposity begins to increase after reaching its lowest point in infancy. The risk of adiposity into adulthood has been associated with the age at which adiposity rebound occurs, and the earlier this occurs the greater the risk (Cole et al., 1995). However, this term is now considered a misnomer as FFM also increases around this time and the term 'BMI rebound' should be used (McCarthy, 2014).

In the UK, the British 1990 (UK90) BMI centile curves are used to define overweight and obesity in children from birth to 23 years, which is based on nationally representative data collected between 1978 and 1990 (Cole et al., 1995). The UK charts have nine centile lines, ranging from 0.4 and 99.6, with the 50th centile representing the median BMI for the sex-specific population at a specified age. The data is smoothed using the LMS method, an advanced statistical tool that accounts for skewness and enables individual standard deviation scores (SDS; also referred to as centile or Z scores) to be determined, which are used to assess the position of the child on the centile chart at the specified age (Cole & Green, 1992; Gatineau & Mathrani, 2011). The Health Survey for England (HSE) 2009 (Aresu et al., 2010), as in previous surveys used the 85th and 95th percentiles as the cut-off points for overweight and obesity respectively. Similarly, the US 2000 CDC (Centers for Disease Control) nationally recommended charts, developed by the US National Center for Health Statistics (NCHS; Kuczmarski et al., 2002), also use the 85th and 95th percentile cut-offs for overweight and obesity respectively for children aged 2-18y.

Use of the 85th and 95th percentile cut-offs has been criticised as arbitrary (Cole et al., 2000), however, these cut-off points continue to be used and are recommended by an Expert Committee comprised of 15 US national health organisations endorsed by a collaboration of the American Medical Association, the Health Resources and Services Administration and the US CDC (Barlow, 2007). For older adolescents, the Expert Committee recommends use of either the adult obesity cut-off of BMI ≥30 or BMI at the 95th percentile; whichever is lower (Barlow, 2007). The HSE cut-off for childhood BMI measures is 15y, with children ≥16y categorized as adults, which would result in potentially fewer adolescents reported as obese than if the

childhood cut-offs were used, as the 95th percentile in the UK90 charts (Cole et al., 1995) used by the HSE is closer to a value of BMI 28. However, as stated above, following NICE (2006) guidelines, the HSE also employs the use of WC as an additional measure of obesity for adults ≥ 16y (Aresu et al., 2010), which may reduce the potential of misclassifying adolescents at the upper limits of the percentile charts (i.e. 85th and 95th percentiles), with BMIs below the adult cut-offs. In clinical settings, when measuring individual children, the 91st and 98th centiles are the recommended cut-offs for overweight and obesity, (Gatineau & Mathrani, 2011) and appear as standard centiles on UK BMI charts.

Internationally two BMI growth reference standards for children are available, the WHO reference curves (de Onis et al., 2007), and centile curves developed by the International Obesity Task Force (IOTF) Childhood Obesity Working group (Cole et al., 2000). The WHO growth reference standards for children and adolescents aged 5-19y (de Onis et al., 2007), were introduced following publication of new standards for infants and children from birth to 5y (WHO, 2006).

These references for 5-19y, were designed to replace the references previously recommended by WHO developed by the NCHS/WHO (de Onis & Habicht, 1996), to take account of the limitations that had been cited in the previous reference charts such as providing assessment measures from age 5y as opposed to 9y, and using advanced statistical methods to produce smooth centile curves, which are also aligned with adult cut-offs for overweight and obesity (Barlow, 2007). In these charts >1SD from the BMI-for-age score represents overweight and >2SD represents obesity (de Onis et al., 2007). Similar statistical methods are also employed in the UK90 charts (Cole et al., 1995; as described above), the CDC 2000 charts (Kuczmarski et al., 2002), and the IOTF charts (Cole et al., 2000).

The latest WHO reference curves are based on the original 1977 NCHS/WHO sample, which is nationally representative of the US population (de Onis et al., 2007), and is the same data set used to construct the CDC 2000 charts (Kuczmarski et al., 2002), except that the WHO dataset excludes the heaviest children, resulting in the BMI levels of the upper percentiles being substantially lower than the CDC

2000 levels, particularly when comparing adolescent BMI levels (Himes, 2009). The IOTF Childhood Obesity Working group (Cole et al., 2000) raised the question of the appropriateness of the WHO recommending the use of the previous 1977 NCHS/WHO charts for international use based on a US reference population. During the development of the latest WHO growth standards (de Onis et al., 2007), an attempt was made to address this matter, however, due to the heterogeneity of methods used for data collection across countries, the WHO reverted back to using the original data set (de Onis et al., 2007). The IOTF charts (Cole et al., 2000) were developed by averaging BMI data across six countries (Brazil, Great Britain, Hong Kong, the Netherlands, and the US). The cut-off points for overweight and obesity for children and adolescents from 2 to 18y were extrapolated back from 18y at adult overweight and obese cut-off points (25kg/m^2 and 30kg/m^2 respectively), and presented in half-year intervals.

With the availability of several different reference charts for determining overweight and obesity for children and adolescents will no doubt cause confusion for health practitioners and researchers when deciding which charts to use, particularly if overweight and obesity outcome varies depending on the charts selected. The NHS National Obesity Observatory (NOO; Gatineau & Mathrani, 2011) guidance, advises that data for overweight and obesity prevalence should only be compared when the same thresholds have been used. Whilst a standardized chart may be useful when making cross-cultural comparisons on obesity and overweight prevalence, it may not be ideal for all populations due to differences in growth patterns and related health risks (Himes, 2009), in particular for certain ethnic groups such as SAs, where as stated previously, there is much evidence of increased health risks for T2DM and CVD at lower BMI and percentage body fat levels (Saxena et al., 2004; Viner et al., 2009). In a recent large UK based study in which adiposity, measured using sum of 4-skinfolds and BIA (Bodystat 1500, Bodystat Ltd, UK) of children aged 9-10y of WE (n= 1345) and SA (n=1523) origin was compared, SA children had higher % body fat and sum of all skinfolds (Nightingale et al., 2011). Furthermore, this study also revealed that SA children had a lower BMI at any given FM value. Similarly, a study conducted in the UK comparing the body composition of SA (n=32 males and 33 females) and WE (n= 31 males and 33 females)

adolescents aged 14-17y, based on skinfold, DXA, and WC measures, revealed that across the range of BMI SDS values, SA adolescents had significantly higher levels of body fat, in particular central adiposity, and significantly lower % LM independent of BMI SDS (Ehtisham et al., 2005). These studies add to the growing body of evidence that BMI is likely to underestimate body fat for this population group, and clearly cannot differentiate between the proportion of lean and fat mass, which is an important measure when determining the health risks associated with body composition.

None of the percentile charts currently in use including the IOTF approved charts (Cole et al., 2000), and the UK90 charts (Cole et al., 1995) are specific to the SA population, and do not account for the increased health risks in this population (Cole et al., 2000; Troiano & Flegal, 1999; Viner et al., 2010). Ethnic specific cut-offs for children and adolescents have been considered, however, an expert group concluded that whilst SA children and adolescents have higher % body fat at a given BMI than white Europeans, this is more apparent in the normal BMI ranges, and thus the current IOTF cut-offs should remain consistent for all populations (Viner et al., 2010). Maintaining this status quo, based on the lower SA adult BMI cut-offs (see section 2.2.1) would lead not only to a proportion of SA adolescents on turning 18y being classified from normal to obese overnight, but children and adolescents with high % body fat levels within the normal BMI range would be classified as 'healthy', whilst potentially being at risk of metabolic syndrome or worse still actually presenting with metabolic syndrome that would likely remain undiagnosed until adulthood, considering that T2DM can be asymptomatic for up to many years (Ehtisham et al., 2000). This would lead potentially to a situation of long-term treatment as opposed to prevention through early detection; which is evident by the much higher prevalence of T2DM in the SA population compared to the white European population (Drake et al., 2002; Ehtisham et al., 2000; Whincup et al., 2010). The expert Group recommended further research into the differences in body composition between SA and white European children to evaluate the potential risks associated with these differences (Viner et al., 2010).

Recent NICE (2013) guidance, has advised in the absence of ethnic-specific growth reference charts, the use of caution when interpreting BMI Z scores based on the UK90 charts for Asian and other minority ethnic groups, in recognition of the recommendations to lower the adult BMI cut-offs for overweight and obesity, within the SA population (see section 2.1.1). However, NICE (2014) does not currently recommend WC as a routine measure for determining obesity in children, but does advise that it may be used for gaining additional information in relation to long-term health risks. In contrast, an IDF Consensus Group (Zimmet et al., 2007), considered WC to be an independent predictor of the metabolic syndrome, and recommended that WC percentile charts be used as an important measure in children and adolescents.

The IDF (Zimmet et al., 2007) has defined use of ≥90th percentile WC cut-off (or adult cut-off if lower) for determining central obesity in children aged 6-<10y, and for diagnosing metabolic syndrome in children and adolescence aged 10 - <16y in addition to two other clinical measures such as raised levels of plasma triglycerides (≥1.7mmol/L) and glucose (≥5.6mmol/L); advising that metabolic syndrome should not be diagnosed in children younger than 10y, although, further investigation was recommended if there was a family history of obesity related diseases such as T2DM and CVD. The 90th percentile cut-off was based on research evidence, which revealed that children at or above this centile were likely to have several risk factors for metabolic syndrome (Zimmet et al., 2007).

The IDF (Zimmet et al., 2007) in its consensus definition of metabolic syndrome (see section 2.1), does not specify actual WC values for children at ≥90th percentile cut-off, but provides as an indication only, one combined ethnicity, and three ethnic- (African-American, European-American, and Mexican-American) and sex-specific WC percentile charts from a nationally representative sample of US children and adolescents (Fernandez et al., 2004). The IDF also recommends further research to develop ethnic- and sex-specific WC ranges. Ethnic-specific WC percentiles are recommended due to evidence of variations in growth as well as the distribution of body fat during early infancy, childhood and adolescence between ethnic groups (Ehtisham et al., 2005; McCarthy, 2006).

In addition to the WC percentile charts produced by Fernandez et al. (2004) for US child and adolescent populations, there are several child and adolescent percentile charts available worldwide (Moreno et al., 1999; Nawarycz et al., 2010; Poh et al., 2011; Zanolli & Morgese, 1996), which are representative of the national populations. In 2001, McCarthy et al. published the first WC percentile charts for British children, based on a representative sample (3,585 boys; 4,770 girls) of the child and adolescent population aged 5-16.9y in 1988, which were comprised of largely WE ethnicity.

As stated in section 2.2 above studies have reported variations between the prevalence of adiposity based on BMI and WC measures, with some studies reporting significant differences between SA and WE children and adolescents (Ehtisham et al., 2005), with others reporting no significant differences (Bhatanagar et al., 1995; Wardle et al., 2006). The Ten Towns Heart Health study found SA children aged 8-11y had significantly greater risk factors for T2DM than WE children, independent of adiposity, based on anthropometric measures such as WC and BMI, with higher levels of fasting insulin and triglyceride, and lower levels of HDL cholesterol (Whincup et al., 2002). This increased risk may be attributed to lower (skeletal muscle mass) SMM within the SA population, as has been identified in previous studies (Lear et al., 2009). Clearly, lower WC and BMI cut-offs recommended for SA adults need to be mirrored in the SA child and adolescent population. In a five-year longitudinal study tracking adiposity (measured by BMI and WC) in Asian (n= 175 girls and 253 boys) and WE (n= 1010 girls and 1597 boys) children from age 11y to 16y, the researchers found that adiposity tracked strongly over time, with children overweight or obese at age 11y likely to remain so at age 16y (Wardle et al., 2006). The researchers concluded that obesity that persists into adulthood is likely to be established during pre-adolescent years. This suggests that for a high-risk group such as SAs, adiposity should be tracked at a much younger age for targeting effective preventative measures.

2.4 Body Composition and the 2,3, and 4 Compartment Models

This section will provide background information on body composition (BC), and how the body is compartmentalised for indirect BC assessment (BCA) into two, three, and four compartment models.

2.4.1 Background

The assessment of individual tissue components would require cadaver analysis, for the direct chemical analysis of the fat and fat-free constituents (Ellis, 2001; McArdle et al., 2007). Other in-vivo methods include magnetic resonance imaging (Goodpaster, 2002) and tissue biopsies, of which the latter would be of limited use in assessment of whole-body composition (Ellis, 2001; McArdle et al., 2007), and apart from the ethical issues and likely difficulties in recruiting willing participants, would be unsuitable in large epidemiological studies. However, direct analysis has helped advance the knowledge of human body composition, such that researchers consider the composition of FM and FFM (i.e. water, protein, and mineral) to be relatively constant, although the ratio of the components of FFM is considered population specific (Ellis, 2000; Heyward & Wagner, 2004; Lohman, 1989; Wells & Fewtrell, 2006). Based on these assumptions researchers have developed mathematical equations to predict body composition from indirect assessment methods (Heyward & Wagner, 2004; Wells et al., 2010).

There is a difference between FFM and LM, as LM includes non-sex-specific essential fat, which is equivalent to approximately 2-3% in males and 5-8% in females (Behnke, 1959 in Lohman, 1992); whereas, FFM excludes all fat including essential fat and is only measurable by direct chemical analysis using extraction methodology (Behnke, 1963; Keys & Brozek, 1953 in Scafoglieri et al., 2011; Lohman, 1992; McArdle et al., 2007; Scafoglieri et al., 2011). Often in research, the terms FFM and LM are used interchangeably, although there is a clear distinction, which leads to confusion, particularly when making comparisons between methods (Heyward & Stolarczyk, 1996; Wang et al., 1992). For clarity therefore, as all the methods described here are based on in-vivo studies, FFM will be considered synonymous with LM.

2.4.2 Body Composition Variations

Whilst the physical properties of fat are relatively constant throughout the lifespan with 0% water and a density of 0.9007g/ml; Formon et al., 1982; Wells et al., 2010), FFM is more variable. At birth the water content of FFM is approximately 80-83%, declining to around 74% by age 16y and to approximately 73% by adulthood (Ellis, 2000; Lohman, 1989; Schoeller, 2005; Wells et al., 2010), which is relatively constant across several adult mammalian species (Wang et al., 1999). Individual variability of ~2% has been reported (Formon et al., 1982; Lohman et al., 2000; Siri, 1961), which could lead to significant errors (~3%) when estimating %FM. In contrast to a decline in TBW with age, bone mineral and protein progressively increases from birth to adulthood, leading to an increase in the density of FFM, from approximately 1.069g/ml at birth, rising to approximately 1.1g/ml in adulthood (Baumgartner, 2005; Formon et al., 1982; Lohman, 1989; Roemmich et al., 1997; Wells et al., 1999). In addition to these age-related variations in FFM density, the density of FFM is also considered population specific, due to proportional variations of its individual components by gender, ethnicity, physical activity history, and level of fitness (Baumgartner et al., 1991; Ellis, 2000; Heyward & Wagner, 2004; Lohman, 1989; Wang et al., 2005; Wells et al., 2010).

To account for the variations in the hydration of FFM during the paediatric years, published reference data for the composition of FFM is commonly used (Wells et al., 2010). In 1982, Formon et al. published data for the 'reference child' from birth to age 10y. In 1989, Lohman combining the data from Formon et al. (1982) published more up to date gender and age-specific reference data for the composition of FFM from age 1-16y, including the %hydration, %BMC, and density (g/ml), together with constants derived from the changing density of FFM and the assumed constant density of FM. These constant densities could be substituted into the Siri (1961) equation to determine %fat (previously described in section 2.4.3a).

More recently, Wells et al. (2010) published paediatric sex-specific reference data for the %hydration and density (kg/L) of FFM from a large sample (n= 261 males & 272 females) of mainly WE (>90%) healthy children and adolescents aged 4-23y based on a 4-C model (see section 2.4.3c), as some of the reference data for children

<10y published by Lohman (1989), was based on the extrapolated measurements from infancy reported by Formon et al. (1982). The new reference data (Wells et al., 2010) was compared to the previously published reference data (Lohman, 1989), which indicated that due to differences in the hydration and densities of FFM, % fat was being underestimated, with a mean bias (Lohman – 4-C value) of -2.1% fat (-3.3% for females & -1.1% for males; P<0.001). According to the more recent reference data (Wells et al., 2010), between the ages of 4-23y the average percentage water content of FFM progressively declines from 76.6% (± 2.4%) to 73.6% (± 1.5%) in males, and from 77.3% (± 2.0%) to 73.7% (± 1.0%) in females, together with a progressive increase in the total FFM density from ~1.082 to 1.1g/ml.

The distribution of FM and FFM is also known to vary with age, pubertal status, between genders, (Baumgartner, 2005; Helba & Binkovitz, 2009; Heyward & Wagner, 2004), as well as between ethnic groups (Dulloo et al., 2010; Ehtisham et al., 2005; Deurenberg et al., 2002; Saxena et al., 2004), physical activity history, and certain disease states (Lohman, 1989). In normal healthy individuals, average %FM ranges from 10-15% at birth, increases to approximately 30% by age 6 months, and gradually declines through early childhood (Baumgartner, 2005). In pre-school aged children, girls have on average 3.8 to 5.5% more body fat than boys (Helba & Binkovitz, 2009). 'Adiposity rebound' or an increase in %FM occurs on average between the ages of 5-8y (Baumgartner, 2005).

The most marked gender differences in both the distribution and % of total FM and FFM are observed during adolescence, with females accruing more gluteal-femoral fat, and males accruing more truncal fat, often described as the 'gynoid' and 'android' phenotype respectively (Baumgartner et al., 2005; Sweeting, 2007). Among males, during adolescence, %FM declines from approximately 17% to 10-13% on average, as FFM rapidly increases (Guo et al., 1997; van der Sluis et al., 2002). Among females, %FM on average steadily increases from 20% to 26% between the ages of 9y through to early adulthood (Guo et al., 1997; van der Sluis et al., 2002).

2.4.3 Body Composition Models

Compartmentalisation of the body's components was first described in 1921 by Czech anthropologist J. Matiega; in this model the human body consisted of: the weight of the skeleton, skeletal muscle, skin plus subcutaneous tissue, and a remainder; the sum of which was equal to body mass. Since this time, due to health risks associated with excess body fat and the difficulty of directly measuring body fat, indirect measurement methods have evolved, which divide the body into 2-4 compartments (Ehtisham et al., 2005; Ellis, 2000; Ellis, 2001; Roche et al., 1996; Wang et al., 2005). Due to the lack of a classification system at the organisational level, Wang et al., (1992) developed a five-level model (Figure 2.1), providing a structural basis for body composition research (Ellis, 2000; Lee & Gallagher, 2008): atomic (I); molecular (II); cellular (III); tissue (IV); and whole body (V), where the total number of components within each level equate to body mass. This section will provide a brief summary of the 2-, 3-, and 4-compartment models commonly used in body composition assessment.

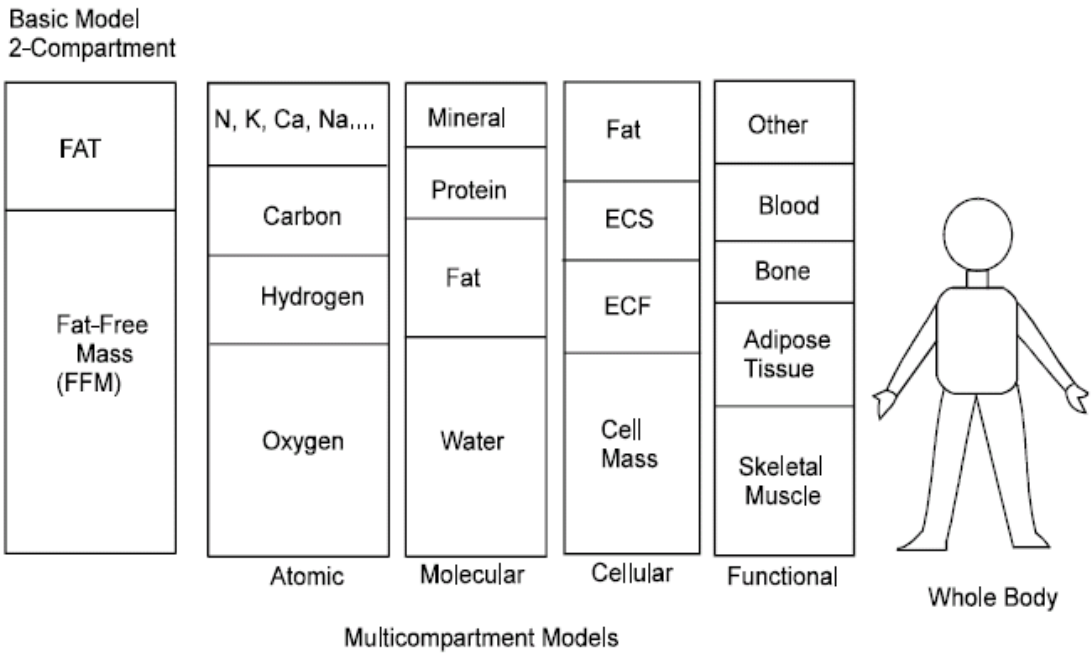


Figure 2.1 Basic two-compartment model and five-level multicompartment model of body composition. ECS, extracellular solids; ECF, extracellular fluid Source copied from: Ellis 2000 (p.652).

2.4.3.1 2-Compartment Models

The classic 2-compartment (2-C) model measuring body mass (BM) or mass and body volume (BV), was pioneered by Bhenke et al. in 1942, and was based on Archimedes' principle that body density (Db) similar to that of an object or substance is a function of the mass and densities of its component parts relative to its volume (Going, 2005), which is defined as its mass per unit volume (i.e. Db = M/V). In this model body BM is divided into FM, and FFM (Ellis, 2000; Ellis, 2001; Heyward & Wagner, 2004; Wells & Fewtrell, 2006), and body volume (BV) is determined by hydrostatic densitometry or underwater weighing (UWW) and more recently by ADP (see section 2.5.3). With assumed constant densities for both FM (0.9g/ml) and FFM (1.1g/ml), led to the development of the reference body, with changes in Db mainly attributed to adiposity (Behnke et al., 1953).

Siri (1956), and Brozek et al (1963) developed prediction equations to determine %FM from Db, which were based on an adult reference body with an assumed Db and %FM, with any difference in the reference Db assumed to be due to %FM (Heyward & Wagner, 2004). This method continues to be used by researchers today, with both equations yielding almost identical results for %FM (Heyward & Wagner, 2004). These equations are presented below, with detailed explanations provided elsewhere of how the densities and proportions of FM and the respective components of FFM are used to derive the constants used in these equations (Heyward & Wagner, 2004; Wang et al., 2005):

%FM = (4.95/Db - 4.50) x 100 (Siri, 1956 equation)
%FM = (4.57/Db - 4.142) x 100 (Brozek et al., 1963 equation)

As explained in section 2.4.2 above, whilst the density of FM is relatively constant, the density of FFM varies with age (particularly during childhood and adolescence), gender, and ethnicity, among other factors, and thus scientists have proposed modifications to these equations based on population-specific conversion formulas, developed from multi-component methods (Going, 2005; Heyward & Wagner, 2004; Lohman, 1989; Wells et al., 2010). Although in the summary of equations provided by Heyward & Wagner (2004), no conversion equation was available specifically for

SA children and adolescents. These conversion equations are considered to provide reasonable estimates of %FM when compared with 4-C model equations (prediction error = 1.9 – 3.4 %FM) at the population level (Heyward & Wagner, 2004). However, greater individual variation has been observed, with 95% limits of agreement (mean ±2SD) ranging from -7.5% to 5% (Roemmich et al., 1997). Other 2-C methods were also emerging at this time, which involved whole body potassium counting (Forbes et al., 1961) and radioactively labelled water (Pace & Rathbun, 1945), which also assumed a constant density for FFM (Ellis, 2000).

Whilst the Siri equation requires a measure of body density, %FM and FFM can also be determined directly from hydrometry (see sections 2.4.3b & 2.6) using the 2-C model formulas (Pace & Rathbun, 1945), based on the age- and gender-specific constant values for TBW as follows:

$$FFM\ (kg) = TBW/C$$

$$\%FM = \{[BM - (TBW/C)]/BM\} \times 100$$

where BM = body mass; TBW = total body water; and C is the age- and gender-specific constant value for TBW = 73.2% for adults.

2.4.3.2 3-Compartment Models

To account for population variability in the hydration of FFM (see section 2.4.2), Siri (1961) developed a 3-C model equation sub-dividing FFM into water and solid (i.e. protein and mineral) fractions, and thus compartmentalising the body into fat, water, and fat-free dry mass (Ellis, 2000; Going, 2005; Heyward & Wagner, 2004; Wells et al., 1999; Wells & Fewtrell, 2006):

$$\%FM = (2.118/Db - 0.78TBW - 1.354)\ 100$$

where Db = body density; and TBW = total body water

This model assumes a constant density for the protein and mineral fractions, and in addition to measuring BM, and BV to determine Db using densitometry; total body water (TBW) is also measured by hydrometry, usually by the isotope (^2H_2O deuterium) dilution method (Ellis, 2000; Heyward & Wagner, 2004; Schoeller, 2005; Wells & Fewtrell, 2006; see section 2.6). Lohman (1986), also developed a 3-C model

using DXA (see section 2.5.2), which divided the body into fat, mineral, and water + protein, to account for variability in the mineral content of FFM. Comparisons between the 4-C model, and the 3-C water and 3-C mineral model revealed much greater errors in the mineral model, which was not recommended for use in children and adolescent populations (Roemmich et al., 1997).

2.4.3.3 4-Compartment Models

The 4-C model further sub-divides body mass into water, bone mineral, fat, and residual mass (including protein, soft tissue minerals and glycogen), with constant densities assumed for the four components (Ellis, 2000; Wang et al., 2005; Wells et al., 1998; Wells & Fewtrell, 2006). In this model bone mineral content (BMC) measured by DXA (Ellis, 2000; Heyward & Wagner, 2004; Wells et al., 1999; Wells & Fewtrell, 2006; see section 2.5.2) is added to the measures in Siri's 3-C model (i.e. BM, BV, and TBW). Although several 4-C equations have been developed (Friedl et al., 1992; Fuller et al., 1992), the resulting estimates of %FM are similar (Wang et al., 2005). The simultaneous equations used to develop 4-C equations are based on the body mass or body mass and body volume models are:

$$BM = FM + TBW + BMC + RM$$

Where BM = body mass; FM = fat mass; TBW = total body water; BMC = bone mineral content; RM = residual mass and units are in kg.

$$BV = FM/0.9007 + TBW/0.99371 + BMC/2.982 + RM/1.404$$

Where BV = body volume; and each component is divided by its assumed constant density in g/ml (Wang et al., 2005).

In the development of paediatric references for the hydration of FFM (see section 2.4.2), Wells et al. (2010) used the following 4-C equation to determine FM in children and adolescents aged 5-20y:

$$FM = (2.747 \times BV) - (0.710 \times TBW) + (1.460 \times BMC) - (2.050 \times BM),$$

(Where FM = fat mass (kg); BV = body volume (l); TBW = total body water (kg); BMC = bone mineral content (kg); and BM = body mass (kg)).

In that study as the majority of the cohort was white, it was recommended that variations in lean tissue composition between ethnic groups be further investigated, although the researchers considered any ethnic differences to be relatively small. The validity and accuracy of any of the equations is dependent on the assumed constants of the densities and proportions of the component fractions to the individual or population to which they are applied (Going, 2005). The 4-C model is considered the most accurate and valid model for estimating body fat for in vivo body composition measurement, as it requires the least assumptions (Going, 2005; Wells & Fewtrell, 2006). However, this model also has an increased risk of measurement error by virtue of the number of different methods involved in determining the body's component parts (Going, 2005; Heyward & Wagner, 2004).

2.5 Body Composition Assessment Methods

Due to the limitations of BMI in assessing adiposity, as detailed in section 2.1 above, researchers have sought to develop valid, practical, and affordable field-based methods that may be more suitable for use in clinical practice, epidemiological studies, and weight management settings (Heyward & Wagner, 2004; Meyer et al., 2011). As explained in section 2.4, the direct chemical analysis of tissues and organs, and a limited number of human cadaver studies (Forbes et al., 1953, 1956; Widdowson et al., 1951) has provided important information on the composition of the human body, and the respective densities of its component parts, which over the last 50y, have been used as a reference for the development of a variety of indirect assessment methods, based on the 2,3, and 4 compartment models (Ellis, 2000; Heyward & Wagner, 2004; Wang et al., 1999; see section 2.2.2). As already mentioned in section 2.4.3, the four most common reference methods used in the

development and evaluation of field-based body composition assessment tools include: hydrodensitometry or UWW, and ADP, used to determine body volume and density; hydrometry using isotope dilution to determine TBW; and DXA used for determining bone mineral density and soft tissue composition (Ellis, 2000; Heyward & Wagner, 2004; Heymsfield et al., 2005).

The 4-C model in which body volume, TBW, and bone mineral are measured separately, is regarded as the 'gold standard' for in-vivo body composition analysis, particularly when the FFM components are variable, as is the case through childhood and adolescence (Heyward & Wagner, 2004; Lee & Gallagher, 2008; Wells & Fewtrell, 2006). However, this method requires the use of much specialised and often costly equipment and expertise, which can be prohibitive in many research settings (Lee & Gallagher, 2008; Wells et al., 2012). In a recent study by Wells et al. (2012), in which the use of a 4-C model was used to determine whole-body fat and fat-free mass SDS for children and adolescents (5-20y), DXA (Lunar Prodigy, GE Medical Systems) showed the highest correlations between these variables and the values obtained by the 4-C method (FM SDS r^2 = 88% & 96%; FFM SDS r^2 = 96% & 94% for males and females respectively). It is important to note however, that the high correlation observed between DXA and the 4-C method maybe limited to the DXA model used in this study, as agreement between DXA models has also been shown to vary, as detailed in section 2.3.3.

Other more complex, costly, and advanced laboratory-based methods include total body potassium counting (TBK) to determine body cell mass, neutron activation analysis (NAA), which measures the body's major elements, and magnetic resonance imaging (MRI), which measures body composition at the tissue level (Ellis, 2001; Heyward & Wagner, 2004; Heymsfield et al., 2005). Over the last two decades, with much technological advancement in BIA, it has become a commonly used and popular field-based method for BC assessment in research and clinical practice, and due to its affordability and ease of use, certain portable models are also available for use by the general population (Dehghan & Merchant, 2008; Ellis, 2001; Heyward & Wagner, 2004; Heymsfield et al., 2005; Kyle et al., 2004; Ward, 2012).

This section will review some of the laboratory- and field-based methods used for indirect BC assessment as explained in chapter aims above. As BIA was the field-based method used in this study, and DXA and ADP were used as reference methods, the main focus of this section will be to review these methods, although for completeness a brief overview of other BC assessment technologies will be provided.

2.5.1 Bioelectrical Impedance Analysis

2.5.1.2 General background

Bioelectrical Impedance Analysis (BIA) is a safe, portable, non-invasive and cost effective tool suitable for BC assessment in large-scale epidemiological studies and clinical and health settings (Ellis, 2000; Ellis, 2001; Haroun et al., 2010; Rush et al., 2006; Ward, 2012). Due to its ease of use, requiring minimal training, high reproducibility and correlation with reference methods, it is the preferred option over anthropometric methods such as skinfold-thickness measurement (Kyle et al., 2004; Mialich et al., 2014). It is however, considered less accurate than the more complex lab-based tools that it is often validated against (Kyle et al., 2004; Sweeting, 2007; Ward, 2012), such as ADP (Lohman & Going, 2006), DXA (Going et al., 2006; Hosking et al., 2006; Kriemler et al., 2009; Lim et al., 2009; Meyer et al., 2011; Pietrobelli et al., 2004; Sluyter et al., 2010), isotope dilution (Haroun et al., 2010; Kehoe et al., 2011; Khan et al., 2012; Liu et al., 2011), underwater weighing (UWW; Jensky-Squires et al., 2008); Magnetic Resonance Imaging (MRI; Bosy-Westphal et al., 2008; Oshima et al., 2010; Wang et al., 2013) and multi-component models in which several techniques are used to determine individual body compartments (Hosking et al., 2006; Wells et al., 2012).

Thomasset (1962) established the basic principles of BIA (Chumlea & Sun, 2005), following his pioneering work, which involved the subcutaneous insertion of two needles to measure the body's electrical conductivity (Heyward & Wagner, 2004; Kyle et al., 2004). The four-surface electrode technique, developed by Hoffer et al. (1969) and Nyboer (1970), soon followed and is used in current practice (Kyle et al., 2004). The first commercially available BIA's were of single-frequency, and were

introduced in the mid-1980s (Lukaski et al., 1985); and multi-frequency analysers became available in the mid-1990s (Kyle et al., 2004).

2.5.1.3 Basic Principles

The basic principles of BIA are based on the 2-C model for predicting FM and FFM (total body mass = FM + FFM; see section 2.2.2), and the relationship between the body's electrical impedance and total body water (TBW; Chumlea & Sun, 2005**;** Heyward & Wagner, 2004; Kyle et al., 2004). Detailed reviews of the basic principles of BIA are provided elsewhere (Chumlea & Sun, 2005**;** Heyward & Wagner, 2004; Kyle et al., 2004). As FFM is comprised largely of water (Figure 2.2), containing the body's electrolytes, it is an excellent conductor of electricity, and fat being anhydrous is a poor electrical conductor (Heyward & Wagner, 2004; Mialich et al., 2014). The electrical impedance of the body is used to determine the volume of TBW from which FFM can be indirectly estimated, based on the assumption that the hydration of FFM is relatively constant (~73%; Figure 2.2; Ellis, 2000; Ellis, 2001; Haroun et al., 2010; Heyward & Wagner, 2004; Rush et al., 2006). Thus FFM could be determined by dividing TBW by 73% (i.e. FFM = TBW/0.73).

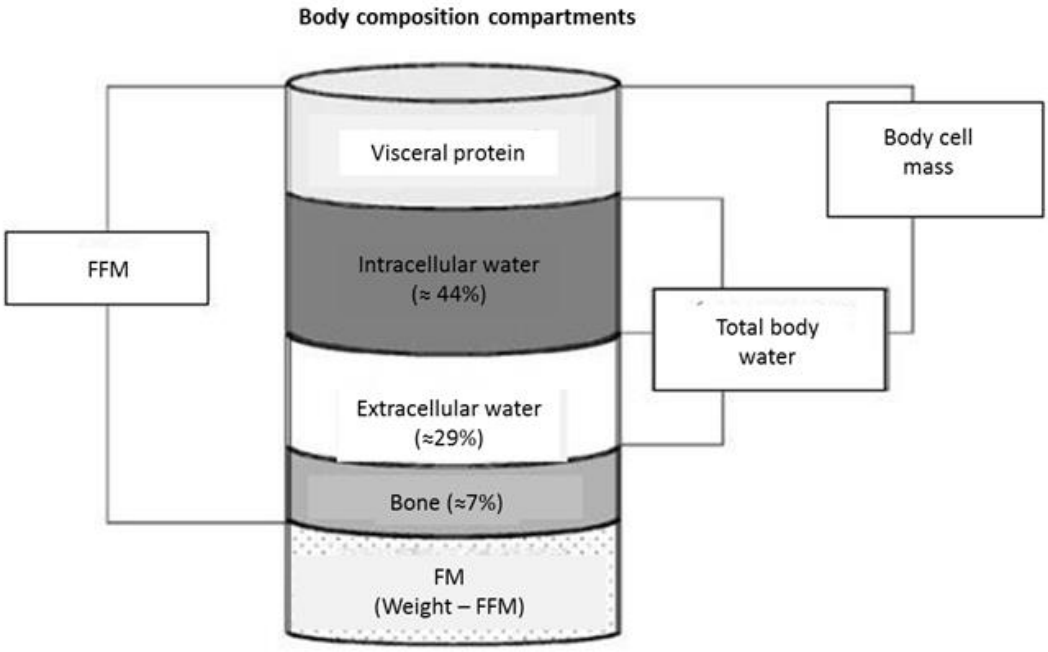


Figure 2.2. Schematic diagram of fat-free mass (FFM), total body water (TBW), intracellular water (ICW), extracellular water (ECW), and body cell mass (BCM). Adapted from Kyle et al., 2004 (p.1231)

In this technique, a low-level, imperceptible electric current is passed through the body at a specified frequency (e.g. 50kHz), and the electrical impedance (Z) or resistance to flow of conducting tissue is measured, from which the volume of TBW (i.e. both intra- and extra-cellular water; ICW and ECW respectively) is determined (Anderson et al., 2012; Chumlea & Sun, 2005; Kyle et al., 2004; Ward, 2012). Impedance is described in the literature as a combination of resistance (R) and reactance (X_C) at a specific frequency (Chumlea & Sun, 2005; Heyward & Wagner, 2004; Kyle et al., 2004), where R is the pure opposition to current flow by TBW and X_C is the opposition to current flow by the cell membrane, described as capacitance or voltage storage (Chumlea & Sun, 2005; Heyward & Wagner, 2004; Kyle et al., 2004). The relationship of impedance with R and X_C is represented by the equation: $Z^2 = R^2 + X_C^2$ (Chumlea & Sun, 2005).

The underlying theory behind BIA is based on Ohm's law and the relationship between electrical resistance and the shape of the conductor (Chumlea and Sun, 2005), where '*resistance (R) of a length of homogeneous conductive material of uniform cross-sectional area is proportional to its length (L) and inversely proportional to its cross-sectional area*' (Kyle et al., 2004, p.1227). Thus, the longer the conductor and smaller the cross-sectional area, the greater the resistance (Chumlea & Sun, 2005). This relationship is described by the following equation:

$$R = \rho L/A$$

Where ρ is the specific resistivity of a homogenous conductor (assumed to be constant); L is the conductor length; and A is the cross-sectional area. By substituting volume (V) into the equation, where V = L x A; and A = V/L then:

$$R = \rho L\,(LV)$$

Which expressed in terms of volume is represented by the equation:

$$V = \rho L^2/R \qquad \text{(Equation 1; Chumlea and Sun, 2005; Kyle et al., 2004).}$$

This equation is also presented with impedance (Z) in place of R as: $V = \rho L^2/Z$ (Haroun et al., 2010; Heyward & Wagner, 2004; Montagnese et al., 2013). For the estimation of TBW the conductive length (L) is considered equivalent to the subject's height (Ht), thus Ht^2/Z or Ht^2/R is proportional to the volume of TBW (Haroun et al., 2010; Kyle et al., 2004). Whilst the human body is clearly not a perfect cylinder of homogeneous conductive material, an empirical relationship is known to exist between the volume of TBW and the impedance index $\rho Ht^2/R$ (Chumlea and Sun, 2005; Kyle et al., 2004). According to Ward (2012), the volume of TBW computed by BIA would be more theoretically accurate, if the resistance of the body segments was measured separately i.e. two arms and legs and the trunk. This would be represented by the equation:

$$V_{total} = 2\,(p\,L^2/R)_{arm} + 2(p\,L^2/R)_{leg} + (p\,L^2/R)_{trunk} \quad \text{(Equation 2)}$$

In reality, the volume or quantity of TBW or FFM is not determined by solving either equation 1 or 2 directly; instead the impedance index ($\rho L^2/R$) is used empirically in regression equations between BIA and body composition measurements obtained from the reference method such as DXA (Chumlea and Sun, 2005; Kyle et al., 2004; Ward, 2012). In its simplest form the regression equation is presented by solving the equation of a straight line, i.e. $y = mx + c$ (where c is the intercept and m is the slope). Replacing y with TBW or FFM (depending on the reference method used) and x with HT^2/Z leads to the following equation:

$$\text{TBW or FFM} = m\,(HT^2/Z) + c \quad \text{(Equation 3)}$$

There is much evidence of variations in electrical conductance of body tissues between population groups (see section 2.2), which has been attributed to differences in hydration levels of FFM, with differences reported between age, sex, and ethnic group, as well as health status (Heyward & Wagner, 2004; Lohman, 1986; Wells et al., 2010), and degree of adiposity (Lee & Gallagher, 2008), with increased TBW and relative ECW observed in obese individuals compared to normal-weight individuals (Pateyjohns et al., 2006). The need to validate BIA models and develop population-specific prediction equations is now widely recognised (Chumlea and

Sun, 2005; Dehghan & Merchant, 2008; Haroun et al., 2010; Kyle et al., 2004; Rush et al., 2006; Ward, 2012), although at an individual level the accuracy of such equations is considered poor, with errors of ±8% fat reported (Wells et al., 1999; Wells & Fewtrell, 2006).

2.5.1.4 BIA models

Several different BIA models are widely available, and in recent years the technology has advanced to offer more valid, reliable, and easier to use methods such as stand-on devices with built-in electrodes (Haroun et al., 2010; McCarthy et al., 2006; Rush et al., 2006; Ward, 2012), than the previous use of simple gel electrodes that required careful placement (Ellis, 2001; Pietrobelli et al., 2004; Ward, 2012). Single-frequency analysers generally employ a frequency of 50 kHz (Chumlea & Sun, 2005; Kyle et al., 2004), as at this frequency the current can pass through both ICW and ECW, and is directly proportional to the volume of TBW (Chumlea and Sun, 2005; Kyle et al., 2004), although there is variability between tissues (Kyle et al., 2004). Multi-frequency analysers are able to differentiate between intra- and extra-cellular water (Chumlea and Sun, 2005**;** Kyle et al., 2004), and are used in clinical and research settings, where in certain disease states such as heart failure and renal disease, hydration status is altered and requires careful monitoring (Kyle et al., 2004). As a single-frequency analyser was used in this study, only this type of analyser will be reviewed here. Detailed reviews of multi-frequency analysers are available elsewhere (Kyle et al., 2004; Ward, 2012).

The two main types of BIA instruments available are bipolar or tetrapolar (Bosy-Westphal et al., 2008; Dehghan & Merchant, 2008). Tetrapolar BIA models are considered more reliable predictors of body composition than the bipolar devices, which is attributed to the greater number of tactile-electrodes, with eight tactile electrodes (hand-to-hand and foot-to-foot) used in the tetrapolar instruments, and four tactile electrodes (hand-to-hand or foot-to-foot) used in the bipolar instruments (Bosy-Westphal et al., 2008; Dehghan & Merchant, 2008; Kriemler et al., 2009). In the bipolar instruments, impedance measurements include the legs and lower trunk (foot-to-foot) or arms and upper trunk (hand-to-hand) only, whereas the tetrapolar instruments incorporate all four limbs and the trunk (Bosy-Westphal et al., 2008; Pietrobelli et al., 2004). The BC-418 8-contact tetrapolar system (Tanita

Corp; Tokyo, Japan) enables whole body and segmental impedance measures to be taken, providing potentially useful information of the distribution of body fat and lean soft tissue (Pietrobelli et al., 2004).

2.5.1.5 Validity, Accuracy and Limitations of BIA

Several studies have assessed and reviewed the validity and accuracy of single frequency BIA in predicting TBW or FFM, with much inconsistency reported in the literature (Bosy-Westphal et al., 2008; Dehghan & Merchant, 2008; Kyle et al., 2004; Meyer et al., 2011). The validity and accuracy of BIA in predicting BC, is attributed to several factors including the parameters used to develop the prediction equation, the criterion method it is validated against, the BIA model employed, as well as the protocols used within the study (Bosy-Westphal et al., 2008; Dehghan & Merchant, 2008; Kyle et al., 2004; Meyer et al., 2011). Use of a standardised protocol has been recommended, to effectively evaluate the validity of studies using BIA for body composition assessment (Kyle et al., 2004).

The BIA models available use proprietary in-built prediction equations validated using a reference method as explained above, that may not be valid for all populations (Dehghan & Merchant, 2008; Kyle et al., 2004; Meyer et al., 2011). Equations developed before 1987 only used HT^2/R or HT^2/Z in the regression analysis to determine TBW or FFM; in later years, additional parameters were included such as gender, age, weight, and anthropometric measures to improve the predictive accuracy of the equations (Kyle et al., 2004). Prediction equations based on the 2-C model are not considered ideal for estimating FFM in children, due to the age related changes in the proportion of water, protein, and mineral components of FFM (Lee & Gallagher, 2008). Use of a multi-component method for validating BIA is considered the most accurate approach (Montagnese et al., 2013; Sun et al., 2003; Wells et al., 2012).

Some of the predictive errors in estimating BC using BIA are attributed to the assumption that the body is a homogenous and uniform conductor of electricity with a constant specific resistivity (p) (Chumlea et al., 1988). However, it is known that intra- and inter-individual differences in specific resistivity are apparent due to differences in tissue composition (Chumlea and Sun, 2005; Kyle et al., 2004), and

proportion of limb lengths, particularly among different ethnic groups (Dehghan & Merchant, 2008). However, in healthy populations that are matched by gender, age, and ethnicity, calibration of BIA against a valid reference method, using appropriate parameters within the prediction equation, would overcome this potential source of error (Ward, 2012).

In segmental analysers, it is considered more accurate to use the segment length for BC estimates of individual body segments (Ward, 2012). However, in practice only standing height is used, which may lead to inaccuracies in segmental measures (Ward, 2012). Whilst measuring segment specific resistivity may be more accurate, particularly in populations where fluid balance is affected by disease state (Kyle et al., 2004), in large epidemiological studies this method would be too onerous and impractical. Certain BIA models also have the option of selecting body type as 'normal' or 'athletic', which would be based on the personal judgement of the operator and may lead to inaccurate results, as significant differences in BC have been observed between the two modes (Dixon & Andreacci, 2011). Due to the inherent problems associated with differences in FFM composition and body geometry, some more recent studies have proposed use of the raw bioimpedance output i.e. Ht^2/Z (Lukaski, 2013; Wells et al., 2012), although this method may be useful for making comparisons with reference data, it would not provide an estimate for %FM or %FFM.

The trunk due to its large cross-sectional area only contributes to approximately 10% of the whole-body impedance measure (Foster & Lukaski, 1996), thus the majority of the impedance measure is related to the FFM of the limbs (Kyle et al., 2004), with the head ignored (Chumlea and Sun, 2005). Due to the complex nature of the trunk, housing all the body's major organs, caution is advised when considering impedance estimates of this region (Chumlea and Sun, 2005). In severely obese subjects, errors may occur in predicting FFM when validated within a sample of mainly non-obese subjects, due to a greater proportion of TBW and body mass accounted for by the trunk (Chumlea & Sun, 2005).

2.5.1.6 BIA and Ethnic-specific body composition equations

Whilst there is much evidence to support the development and use of population-specific BIA equations, there is also evidence of variations between the models used, with tetrapolar devices demonstrating better agreement between reference methods such as DXA and MRI, than bipolar devices (Bosy-Westphal et al., 2008; Kriemler et al., 2009). The validation study detailed in chapter 4 of this thesis used a tetrapolar BIA device (Tanita BC418-MA) to develop a FFM prediction equation for the SA child and adolescent population; therefore, this section of the literature review will focus on BIA validation studies using both bipolar and tetrapolar devices that have developed ethnic specific prediction equations specifically for the SA child and/or adolescent population, or have included SAs within the sample population. Any prediction equations that have been developed for the same population group used within this study will also be assessed for predictive accuracy.

A literature search revealed three studies conducted on SA children and adolescents using BIA, that published ethnic-specific prediction equations for determining FFM or TBW (Haroun et al., 2010; Khan et al., 2012; Sluyter et al., 2010). In both the studies conducted by Haroun et al. (2010), and Khan et al. (2012) the Tanita (TBF-300) leg-leg BIA model was used, and the reference method was deuterium oxide (D_2O) dilution (see chapter 2 General Methods for details on this technique), from which TBW was estimated, whilst the study conducted by Sluyter et al (2010) used the same Tanita BC418-MA model, as proposed in the BIA validation study in this thesis, with DXA as the reference method.

In the study conducted by Haroun et al. (2010), the sample population recruited from two East London schools, comprised three distinct ethnic groups of white, black, and Asian (Indian, Pakistani, and Bangladeshi) healthy adolescents (aged 11-15y). FFM was determined based on sex-specific hydration equations from an on-going and unpublished research project, which factored in age-related changes in FFM hydration. The equations for calculating hydration of FFM were: hydration = 78.176 – (0.237 x age), and 79.797 – (0.385 x age) for males and females respectively.

In the study conducted by Khan et al. (2012), which involved Bangladeshi children aged 4-10y, from a poor rural region of Bangladesh, the authors stated that the sex- and age-related hydration of FFM was determined based on published reference values by Lohman (1989, 1992), and ranged from 76.2% to 78.3%. The approach used in determining FFM differed between the studies, with Haroun et al. (2010) developing sex-specific equations to determine TBW with D_2O as the dependent variable, and Ht^2/impedance and ethnicity as the independent predictors in the regression model. FFM was then determined using the sex-specific hydration equations as previously mentioned. Khan et al. (2012), on the other hand, converted TBW estimates from D_2O to estimate FFM first, which was then used as the dependent variable, to develop a prediction equation, with sex, Wt, age, and Ht^2/impedance as the independent predictors in the regression model. This equation however, did not ascribe any numerical value to sex, and was thus difficult to evaluate.

The sample population in the study conducted by Sluyter et al. (2010), were healthy adolescents aged 12-19y from a multi-ethnic background, including Asian (n = 61 male and 59 female) and WE (n = 37 male and 54 female). The Asian group were of mixed ethnicities, and whilst the majority (n= 49 male and 41 female) were Indian, the remainder were from 10 different countries including Vietnam, Cambodia, China, with only 2 Pakistani females, and no Bangladeshis in the sample. Having a mixed Asian sample may have impacted the validity of the prediction equation specifically for SA adolescents. In that study BIA compared to DXA on average significantly ($P<0.001$) underestimated %FM by 2.84% (SD= 4.70%), with a

tendency to overestimate %FM in lean subjects and underestimate %FM in subjects with high adiposity. This finding has also been observed in other studies (Haroun et al., 2010; Pietrobelli et al., 2004; Prins et al., 2008).

Due to the uncertainties with the differing equations, and the high standard error of estimates (SEE) in absolute values, Wells et al. (2012) proposed comparing raw outputs of H^2/impedance in BIA with the reference method used, in SDS format. This approach enables comparisons to be made between different body composition assessment techniques, to determine whether FM or FFM percentile rankings from one technique, e.g. DXA or the 4-C model, are consistent with BIA, although this would prevent direct comparisons of absolute values to be made.

In a recent study, Montagnese et al. (2013) assessed the validity of a single BIA equation to determine FFM on a heterogeneous sample of 547 (240 males) children and adolescents (4-24y), using a 4-C model and a tetrapolar BIA instrument (Tanita BC-418MA). Step-wise multiple regression analysis was used to determine whether age (separated into 6 distinct age groups), gender, pubertal stage (Tanner stages 1-5), and nutritional status (BMI SDS separated into non-overweight <1.04; overweight 1.04 - 1.639; & obese > 1.64) resulted in significantly different values for the slope (m) and intercept (c) in the regression equation (equation 3 above) for determining FFM from Ht^2/Z.

In that study, age and pubertal status exerted the greatest effect on the regression equation with significantly different values for the slope (m) and intercept (c) in the age groups 4-7y and 16-19y. These differences were attributed to the age-related changes in body proportions, as limb lengths increase in relation to trunk. It was concluded that whilst HT^2/Z was a strong predictor of LM ($r^2 = 0.953$, SEE = 2.9kg), no single equation could be a valid predictor of LM or TBW across a wide age range of children and adolescents, due to significant differences in the slope (m) and intercept (c) values. However, ethnicity was not explored in this study, with 80% of the sample reported to be European and 20% non-European from a variety of different ethnic groups. This omission may have impacted the outcome of the regression model, given that ethnic differences in body composition are widely

recognised in the literature, and many studies have published ethnic-specific prediction equations.

2.5.2 Dual-energy X-ray absorptiometry

2.5.2.1 General Background

Dual-energy X-ray absorptiometry (DXA) was originally introduced to measure bone mineral density (BMD; Laskey, 1996; Tothill et al., 2001), (Laskey, 1996); however, it is now one of the most widely used methods for whole- and regional-body composition analysis as it is non-invasive, requires little subject cooperation, with newer models offering fast scan times (lasting ~ 5 minutes for a whole-body scan; Bonnick, 2010), is regarded as safe due to its extremely low-dose radiation, and is relatively simple to use (Helba & Binkovitz, 2009; Heyward & Wagner, 2004; Laskey, 1996; Plank, 2005). Whilst DXA provides composition estimates for 3 body components (i.e. BMC, FM, and LTM), it is in fact a 2-C model, based on two separate sets of equations (Ellis, 2000; Testolin et al., 2000). In DXA, soft tissue FFM is often described as lean tissue mass (LTM), and the terms are often used interchangeably (Kelly et al., 1998; Wells et al., 2010). For the sake of consistency and to avoid confusion, FFM without bone will be described as lean tissue mass (LTM), and FFM will represent BMC and LTM.

There are three commercial manufacturers of DXA scanners and whilst there are slight differences between the various DXA models, all have similar features i.e. a scanning table, a dual-energy X-ray source with a detector linked to a computer system for generating scanned images, and body composition data (Helba & Binkovitz, 2009; Heyward & Wagner, 2004; Laskey, 1996; Plank, 2005). The software system processes and analyses the individual X-ray absorption measures, presenting quantitative data for FM, LTM, and BMC in grams, and BMD in g/cm^2. Total body mass is calculated as the sum of all three parts (i.e. FM + LTM + BMC) (Korth et al., 2007; Lohman et al., 2000).

DXA systems employ two different scanning technologies, namely the 'pencil-beam' and 'fan-array' scanner (Bonnick, 2010; Laskey, 1996; Plank, 2005). The fan-array systems compared to the pencil-beam scanners have a significantly reduced scan

time, with improved precision and image-resolution (Bonnick, 2010; Laskey, 1996). However, the radiation dose in the fan-array scanners is 2-5 times greater than the pencil-beam devices (Laskey, 1996). The more recent pencil-beam scanners such as the Norland XR-800 also offer much faster scan times (Plank, 2005), with very low dose radiation.

The radiation dose used in DXA scans is extremely low and is estimated to be similar to one day's exposure to natural background radiation (Helba & Binkovitz, 2009; Bonnick, 2010). In the pencil-beam devices, such as the Norland XR-800 bone densitometer the scan dose for a whole body scan is <0.1mrem (or1 μSv), which is approximately equivalent to 1/6 of normal background radiation (see Appendix A1). The department of Medical Physics at the Royal Devon and Exeter Hospital, and the Radiography department at Exeter University conducted a 'DEXA risk and benefit assessment' (Ward & Knapp, 2007); a copy of the tables produced in this report comparing DXA doses with other common activities in which radiation exposure occurs are presented in Appendix A2, to put DXA scans into context with other common exposures to radiation. For example, travelling 30 miles by car would expose an individual to more than 10 times the dose of radiation than a whole body DXA scan using the Norland XR-800 scanner.

2.5.2.3 Basic Principles

The DXA system generates an X-ray beam at two photon energies, and divides the body into a series of pixels (Plank, 2005), which on penetrating the body tissues are attenuated; the degree of attenuation varies by tissue density, thickness, and chemical composition (Bonnick, 2010; Heyward & Wagner, 2004; Mazess et al., 1990), with denser tissues such as bone absorbing the greatest photon energy compared to soft tissue (Plank, 2005). This difference in the attenuation of the X-ray photon energies within each pixel is measured by a detector and is processed by the software to create the X-ray image (Bonnick, 2010; Laskey, 1996), which is used to calculate the in vivo tissue composition (Laskey, 1996; Plank, 2005; Scafoglieri et al., 2011).

As the DXA scan involves two photon energies, in theory only two components can be identified by the differential absorption within each pixel (Laskey, 1996; Plank, 2005). The absorption of the low-energy photons relative to the high-energy photons is expressed as a ratio (R) (Heyward & Wagner, 2004; Laskey, 1996). Calibration phantoms of known composition are used to determine the attenuation coefficients for bone, fat and fat-free soft tissue (Heymsfield et al., 1989; Mazess et al., 1990). The R-values from the pixels containing the soft tissue fraction are used to determine the amount of fat and fat-free mass, which makes up approximately 60% of the body (Ellis, 2000; Laskey, 1996; Lohman & Chen, 2005). The tissue composition of the remainder of the scan (containing BM and LTM) is determined by extrapolation from the LTM (Ellis, 2000; Laskey, 1996; Lohman & Chen, 2005; Plank, 2005). BCA of the 3-components is thus based on repeated data acquisition of the whole-body scan, with R-values for each component assumed to be constant (Laskey, 1996; Pietrobelli et al., 1996; Testolin et al., 2000). The algorithms used for determining the attenuation coefficients and R-values, to determine mass values for BMC, FM, and LTM, are provided in a review by Pietrobelli et al. (1996).

2.5.2.4 Accuracy & Precision of DXA

DXA technology is being increasingly used as a reference method for BCA, and has been the subject of several reviews and validation studies to assess its viability as an acceptable reference method based on its precision (or repeatability) and accuracy (or validity) (Lohman et al., 2000; Plank, 2005; Scafoglieri et al., 2011). Short-term precision is evaluated by repeated measures of each subject (within and between days), and is based on intra-individual variation of subject scans, often reported as the percentage coefficient of variation (%CV) together with the standard deviation. Long-term precision is based on scans repeated over several months (Lohman & Chen, 2005).

For DXA to be considered an accurate criterion method for BCA, the standard error of estimate (SEE) should be less than 3% (Lohman & Chen, 2005). Errors of 3-4% have limited validity, and >4% would render DXA too variable for use as a reference method (Lohman & Chen, 2005). DXA studies have reported good to excellent short- and long-term precision, with %CVs ranging from 1.4-2% for FM, and 1.1-1.5% for LTM (Marguiles et al., 2005; Mazess et al., 1990; Njeh et al., 1997). DXA accuracy is

more complex involving direct comparison studies between chemical analyses of animal carcasses, (Helba & Binkovitz, 2009; Plank 2005), with no validation studies involving cadaver dissection (Plank, 2005; Wells et al., 2010), and relatively few involving animal carcass dissection (Scafoglieri et al., 2011), which are often described as the gold standard techniques.

Indirect assessment methods using multicomponent models, in particular the 4-component (4-C) model, are more commonly used as the criterion method to assess the accuracy of DXA in human BCA (Lohman, 2000; Plank, 2005; Wells et al., 2010). In the 4-C model body fat is estimated from three different assessment methods for determining body density, total body water, and bone mineral content, namely: densitometry (UWW or ADP), hydrometry (usually deuterium dilution), and DXA respectively (Plank, 2005; Wells et al., 2010).

Early studies of DXA accuracy reported poor results (Ellis et al., 1994). Reviews of studies on the more recent DXA scanners have been variable (Lohman, 2000; Plank, 2005). Some studies have reported excellent accuracy, when compared to chemical analysis of animal carcasses, with variations of approximately 1.0-2.9% for FM, and 3% for LTM (Black et al., 2001; Svendsen et al., 1993), whilst other studies have reported more variable outcomes (Brunton et al., 1993; Ellis et al., 1994; Scafoglieri et al., 2011). These differences have been attributed to a number of factors including: (i) the R-value constancy assumptions for bone mineral, fat, and LTM regardless of subject population (Helba & Binkovitz, 2009; Lohman et al., 2000; Pietrobelli et al., 1996); (ii) variations in tissue depths (Jebb et al., 1993; Laskey et al., 1992; Pietrobelli et al., 1996); (iii) differences in scale Wt versus DXA sum of parts (Friedl et al., 1994; Nelson et al., 1996); (iv) methodological differences between validation studies, and technological (software and hardware) differences between manufacturers of DXA scanners (Clasey et al., 1999; Heyward & Wagner, 2004; Lohman, 2000; Plank, 2005; Scafoglieri et al., 2011; Testolin et al., 2000). The impact of these differences on the accuracy and precision of DXA are explained in detail elsewhere (Helba & Binkovitz, 2009; Heyward & Wagner, 2004; Plank, 2005; Testolin et al., 2000).

2.5.3 Densitometry

Densitometry refers to the measurement body density (Db), determined by measuring body volume based on the 2-C model (see section 2.2.3). Traditionally, this method involved hydrostatic weighing (HW; Ellis, 2001; McCrory et al., 1995; Wells & Fewtrell, 2006), which was developed in the early 1940s by Behnke et al. (1942). Measurement of body volume by HW is considered impractical for field use and is considered particularly unsuitable for children, as it requires complete underwater submersion of the subject with the maximal exhalation of air to account for residual lung volume, and tests have to be done repeatedly to ensure reliability (Ellis, 2001; McCrory et al., 1995).

HW uses the Archimedes' principle for the determination of body volume, whereby an object's specific gravity is determined by the Wt of the object in air divided by its loss of Wt in water, from which the object's density (D= mass/ volume) can be determined (McArdle et al., 2007). As explained in section 2.2.3 scientists Siri (1956) and Brozek (1963) incorporated these densities into equations to determine %FM, which yielded almost identical results (Heyward & Wagner, 2004; Wang et al., 2005). To account for population variations in the density of FFM (see section 2.2.2), 3- and 4-C model equations were developed (Ellis, 2000).

ADP is based on the 2-C method (body mass = FM + FFM) designed to measure body volume, which has evolved from the HW technique (Ellis, 2000; Going, 2005; Heyward & Wagner, 2004;). The Bod Pod® Body Composition System (Life Measurement Instruments, Concord, CA) is a relatively recent air displacement plethysmograph, which is considered highly reliable and valid as a field-based tool for measurement of body volume, having been validated against HW (McCrory et al., 1995). The Bod Pod is constructed of fibreglass and consists of two chambers (Figure2.3) for determining body volume, connected to a computer with the appropriate measurement software, and electronic scales for the measurement of body mass to the nearest ± 5g. The 450l volume front chamber has a moulded seat and a large acrylic window, and an electromagnetic door with an internally operable release button. The rear chamber houses the necessary instrumentation, which is described in more detail elsewhere. The volume of the subject is determined by the

displacement or reduction of air in the front chamber. The volume of air in the lungs is deducted from the measured volume to determine the body volume. The pulmonary gas volume can either be measured directly by connecting the subject to a breathing circuit, or can be predicted by the computerised software (McCrory et al., 1995).

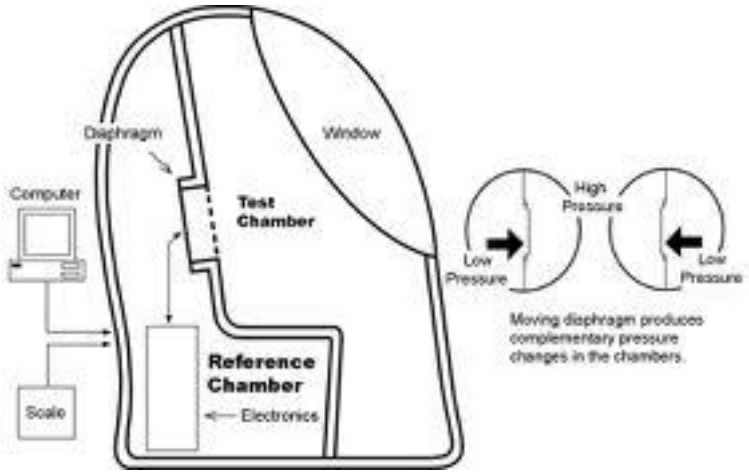


Figure 2.3 – Diagram of how the Bod Pod® measures body volume

Percentage body fat is determined by use of the Siri equation, and similar to HW its accuracy as a 2-C method, to determine %FM based on this equation has been questioned, due to the well documented population variations in FFM density, particularly during childhood, and between ethnic groups (Ellis, 2000; Going, 2005). Whilst the Bod Pod is considered a valid, reliable, relatively simple, and non-invasive method for measuring body volume and density, it is more suited to laboratory- or clinic-based assessment, rather than for large-scale epidemiological studies.

2.5.4 Hydrometry

Hydrometry is another commonly used in vivo 2-C technique (where BM = FM + FFM) in BC assessment, which determines TBW from which FFM can be estimated, based on the assumption that the hydration of FFM is relatively constant for a given population (Ellis, 2000; Lohman, 1989; Schoeller, 2005; Wang et al., 1999). This method is based on the principle that the volume of the solvent (in this case TBW) is equal to the ratio of the dose of the solute (isotopic tracer) divided by its concentration within the solvent (Edelman et al., 1952).

2.5.5 Alternative Assessment Methods

Other more complex laboratory-based body composition assessment methods are available e.g.: (i) whole-body potassium counting; (ii) neutron activation analysis (NAA); (iii) computerised tomography (CT); and (iv) magnetic resonance imaging (MRI), but due to the cost and level of expertise required are not commonly used (Ellis, 2000). Furthermore, some of these methods do not provide improved accuracy or reliability, and lack reference data (Wells & Fewtrell, 2006).

In whole-body potassium counting, the radioactive isotope of potassium (^{40}K) is counted and converted to a total body potassium (TBK) value (Ellis, 2000; Ellis, 2005; Heyward & Wagner, 2004), and in BCA it is used to determine body cell mass (BCM; Ellis, 2005) and FFM (Ellis, 2000) based on the constancy assumption. In NAA the body is exposed to a neutron field, which captures the body's atoms and some of these are converted to radioactive isotopes of the original atoms, emitting gamma rays that can be measured in a similar way to ^{40}K (Ellis, 2000). CT scanning is an X-ray imaging method, similar to DXA that involves the transmission of X-rays, which rotate around the whole body in a fan-shaped beam (Ellis, 2000; Ross & Janssen, 2005). Whilst this method has been shown to provide excellent precision and accuracy (<1% error) in determining body composition (Ellis, 2000), the cost and radiation dose used in this procedure, has limited its use in BCA (Ellis, 2000; Heyward & Wagner, 2004; Ross & Janssen, 2005). As with DXA and CT scanning, MRI also provides in-vivo images of the body's components at the tissue level (Ellis, 2000; Heyward & Wagner, 2004; Ross & Janssen, 2005), although this technology uses magnetic and radiofrequency fields rather than radiation to generate its images (Ellis, 2000; Heyward & Wagner, 2004; Ross & Janssen, 2005). Whilst MRI is considered very useful in BCA, its use is restricted due to its high cost, limited availability, as well as the time required for the procedure (Ellis, 2000; Heyward & Wagner, 2004; Ross & Janssen, 2005). More detailed information on these reference methods is available elsewhere (Ellis, 2000; Heymsfield et al., 2005; Heyward & Wagner, 2004; Wells & Fewtrell, 2006).

2.6 Stature and Body Proportionality – leg length and sitting height

Anthropometry, particularly during childhood, is widely used in clinical and public health work as a measure of growth and as a predictor of health (Cameron et al.,

1981). Whilst short stature in adulthood is associated with an increased risk of obesity (Guerrero-Igea et al., 2001; Gunnell et al., 2003; Lawlor et al., 2002) and metabolic syndrome (Guerrero-Igea et al., 2001; Lawlor et al., 2002; Smith et al., 2001), taller for age children have been shown to have a greater propensity to obesity (Baker et al., 2007; Bosy-Westphal et al., 2009; Buchan et al., 2007; Demerath et al., 2009; Himes et al., 1986; Lazarus et al., 1996). The relationship with taller stature and obesity in childhood is associated with advanced sexual maturity among girls (Adair & Gordon-Larsen, 2001; Freedman et al., 2003), with some evidence of advanced skeletal maturation between both sexes (Demerath et al., 2009). Although, the latter association may be more strongly related to linear growth, as the link to advanced skeletal maturation in obesity is not so strong (Akridge et al. 2007). From birth to the age of 7 years, the legs grow faster than other body segments (Asao et al., 2006; Bogin & Valera-Silva, 2010). Subischial leg length continues to increase rapidly during the prepubertal years, with a more gradual decline in the growth of leg length among boys than girls (Dangour et al., 2002).

Shorter leg length (LL) and particularly shorter leg length relative to Ht, identified in some studies as a lower leg length to height ratio (LLHR; LL/Ht) or a higher sitting height ratio (SHR; SH/Ht), is a marker of an increased risk for non-communicable diseases such as T2DM, obesity, and CVD, in adulthood (Asao et al., 2006; Bogin & Varela-Silva, 2010; Pliakas & McCarthy, 2010; Hales & Barker, 2001; Smith et al., 2001; Wadsworth et al., 2002). Recent research has also revealed that taller for age children have a lower LLHR (Pliakas & McCarthy, 2010), which adds to the growing body of evidence that LLHR is an important contributor to the already established risk factors to adult health.

Whilst the adult size and proportionality of body segments has been attributed to an interaction between genetics and the environment, research has indicated that the environment exerts a greater influence on body proportions, in particular LL, than genes (Asao et al., 2006; Bogin & Varela-Silva, 2010; Dangour et al., 2002; Tanner et al., 1982; Wadsworth et al., 2002). Growth, with respect to LL and LLHR or SHR during infancy and childhood is highly plastic (i.e. strongly influenced by environmental factors), with adverse environmental conditions such as poor foetal

or infant nutrition (Asao et al., 2006; Wadsworth et al., 2002), and low socioeconomic status directly affecting stature and limb length (Bogin & Varela-Silva, 2010; Wadsworth et al., 2002). Evidence of the plasticity of LL and LLHR or SHR and the associated increased health risks, underpins contemporary hypotheses such as the thrifty phenotype hypothesis (TPH; Hales & Barker, 2001).

The TPH, originally developed in 1991, states that when foetal or early infant nutrition is poor, the body directs nutrition more favourably towards the growth and development of vital organs such as the brain and those in the trunk; leading to reduced growth in terms of birth weight or overall body size leading to shorter LL and particularly a lower LLHR in adolescence and adulthood (Bogin & Valera-Silva, 2010; Hales & Barker, 2001).

Among infants born in such adverse environmental conditions, improved nutrition during childhood can lead to a rapid increase in weight and stature, often described as 'catch up growth', with those children experiencing rapid weight gain showing an increased risk of developing T2DM in adulthood (Hales & Barker, 2001). A positive association has been found with final leg length and breast-fed babies (Martin et al., 2002; Wadsworth et al., 2002), and energy consumption during early childhood years (Wadsworth et al., 2002). Considering the paradox that taller for age children and shorter adults have an increased risk of obesity, T2DM, and CVD (Demerath et al., 2009; Gunnell et al., 2003; Lawlor et al., 2002); longitudinal studies comparing the LLHR or SHR of adults undergoing a rapid increase in weight and stature during early childhood, against those experiencing a slower increase in weight and stature, may provide an explanation for this paradox, alongside other measures of risks associated with T2DM.

Whilst there is stronger evidence for environmental influences on body size and proportionality, than genes, there is also much evidence of significant variations between population groups throughout the world (Dietz et al., 1989; Eveleth & Tanner, 1990; Fredriks et al., 2000; Tanner et al., 1982), although, research has shown that populations migrating to environments with improved nutrition and health, leads to increased stature, in particular relative leg length (Akram &

Agboatwala, 1991; Bogin et al., 2002; Hamill et al., 1979). For instance, Bogin et al. (2002), conducted a study comparing the Ht, SH, and SHR of children (n=431; aged 5-12y) from Maya families that had migrated in large numbers to the US between the late 1970s to early 1990s, with indigenous Maya children living in Guatemala, and found that the Maya American children were significantly taller (+11.54cm), with longer legs (+6.83cm), than their Guatemalan counterparts. Similarly, longitudinal studies have shown that children of Pakistani origin from more affluent backgrounds living in the USA have very similar growth patterns to the general population, when compared to the growth curves of the National Centre for Health Statistics (NCHS; Akram & Agboatwala, 1991; Hamill et al., 1979).

Eveleth and Tanner (1990), in their collation and interpretation of studies on the worldwide variation of human growth, reported that for overall stature, Indian boys aged 2-10y from a more affluent background had similar heights to WE populations, whilst the national average of Indian children was much lower. However, peak height velocity or adolescent growth spurt, whilst it occurred at a similar age for Indians and Europeans, (11-12y, and 13-14y for girls and boys respectively), the rate of growth and overall adult height among Indians was lower, even among the affluent Indians. They also reported that the SH for Indian children was lower than for Europeans, although the relative SH was similar. A more recent review examined variations in pre- and post-pubertal growth of children and adolescents from high SES groups, across 53 population groups covering all major continents, to the National Centre for Health Statistics/World Health Organisation (NCHS/WHO) references (Haas & Campirano, 2006). In this review it was reported that pre-adolescent Ht was more similar across population groups worldwide, with non-Europeans approximately 5cm below the reference and northern Europeans 5cm above the reference. However, after puberty, the divergence in Ht increased. It was concluded that a single reference standard might only be appropriate for pre-adolescent Ht.

In addition to health and nutrition influencing secular changes to stature and limb length, there is some evidence to support the theory that climatic conditions over several generations may underlie the genetic variations in stature, particularly

relative leg length (Bogin & Valera-Silva, 2010), with populations from colder climates having relatively shorter limbs in relation to Ht to minimise heat loss, than those from warmer climates (Ruff, 2002, Eveleth & Tanner, 1990). Studies in the USA, including the NHANES III 1988-1994 survey of adults aged 20-49y (Bogin & Valera-Silva, 2008) have shown that black adults and youths, with the same stature as their white counterparts, have relatively longer limbs (Hamill et al., 1973; Krogman, 1970). In a prospective study the early phenotypic development of Indian children (n= 663) from Mysore, India, measured at birth, one and four years, was compared to UK and Dutch growth standards (Krishnaveni et al., 2005). Compared to the UK children, the Indian children were smaller in size for all measurements, at birth and one year, although the crown-heel length and sub-scapular skinfold thickness revealed the least deficit, and at 4 years the Indian children remained considerably smaller, apart from sub-scapular skinfold which was larger, and the Ht difference was also less pronounced than other measures, however LL comparisons were not available for UK children at 4 years. Similarly, compared to the Dutch children at 1 and 4 years the Indian children were smaller for all measures apart from sub-scapular skinfold which was greater, with a much lower crown-rump length (~-2SD) or SH (~-2.7SD) at 1y and 4y respectively, however, LL was close to the average Dutch standards, indicating relatively longer legs.

References representing the relevant measures for a specific population are commonly employed to make comparisons between ethnic groups. However, due to evidence of secular changes in childhood growth patterns, leading to increased stature and weight (Cole et al., 1998; Freeman et al., 1995); and differences between ethnic groups (Chinn et al., 1998; Eveleth & Tanner, 1990; Rona & Chinn, 1986; Saxena et al., 2004), for the purposes of validity, it has been repeatedly stated that such reference standards need to be up to date, and representative of the specific population group (Dangour et al., 2002; Freeman et al., 1995; Kelly et al., 1997).

To account for secular changes in Ht, which have been attributed to a greater increase in LL over SH, Dangour et al. (2002), updated the Tanner and Whitehouse reference curves for SH and LL for British children, that were constructed in the 1970s (Tanner & Whitehouse, 1978), using a convenience sample of 1424 boys and

1208 girls age under 25y, from Southeast England, measured between 1995-1996. Non-white children were excluded from the analysis due to assumed ethnic differences. Subischial LL was determined by subtracting SH from Ht. The analysis did not include relative LL, such as the SHR or LLHR, making such comparisons difficult, with other population groups, where stature may differ (Bogin & Valera-Silva, 2010). Eveleth and Tanner (1990) published data for comparing population differences in relative LL based on the SHR, which covers four continents (Australia, Africa, Europe, and Asia) across the world. However, with evidence of secular changes in overall stature and RLL, associated with the environment and socioeconomic status, suggests that such comparisons should be interpreted with caution and ethnic-specific, up to date references should be referred to (Dangour et al., 2002; Freeman et al., 1995; Kelly et al., 1997).

2.7 Summary

In summary, this literature review provides evidence of the greater health risks for MetS associated with overweight and obesity for the SA child and adolescent population, compared to their WE counterparts. The research shows that the current BMI thresholds for defining overweight and obesity are too simplistic and are likely to underestimate the health risks associated with excess adiposity for this population group, and lowering these thresholds in line with the SA adult cut-offs may go some way towards identifying more individuals potentially at risk of MetS.

WC is also recommended by the IDF as an important tool for diagnosing MetS in children; although measuring WC is not currently recommended by NICE for children, its use has been advised for determining further information on long-term health risks. Lower WC cut-offs are used for determining overweight and obesity in SA adults compared to WEs, and India has introduced WC cut-offs as 'Action points' for lifestyle change or further medical assessment in relation to T2DM and CVD. Nationally representative percentile reference curves for BMI are used for assessing children and adolescents, with specific cut-offs used for defining individuals as underweight, normal weight, overweight, or obese. The UK90 percentile charts are based on WE populations, with no similar reference charts for SAs, and NICE have advised use of caution when interpreting the UK90 BMI charts for non-WE

ethnicities. WC, %FM, and SMM reference curves have also been developed for the UK WE population. Similar reference charts are not available for the SA population.

Research shows that more direct methods of determining body composition in large-scale public health settings would be a preferable adjunct to BMI and WC. BIA has been identified as a potential tool for BCA, due to improvements in its accuracy, ease of use, portability, and cost effectiveness. The BC418-MA, a portable tetrapolar BIA instrument has been found to be a suitable field-based model for measuring BC as it provides whole-body estimates of %FM as well as appendicular FFM or SMM (FFM_a/SMM_a), which enables more detailed BCA, as reduced FFM_a or SMM_a is a further indicator of health risk associated with MetS. The proprietary in-built equations used for determining BC in the BC418-MA are based on WE populations, and have been shown to underestimate FM (and hence overestimate FFM) in SA populations. BIA BC prediction equations are developed by using regression analyses with a reference method such DXA, ADP, or hydrometry. Only one study was identified in this literature review that had developed a FFM prediction equation for the SA adolescent population using the BC-418MA model. No studies were found that had determined SMM_a for the SA child and adolescent population.

A lower LLHR or RLL has also been shown to be an indicator of health-risk associated with obesity-related diseases such as MetS, however, no contemporary UK data on the SA child and adolescent RLL was found in this literature review. Some studies revealed that both SA child and adolescent populations had similar or longer RLL. There was also no available portable tool for determining RLL by measuring SH.

2.8 Thesis Aims

The aims of this thesis (limited to the UK SA child and adolescent population) were to:

1. Develop BIA prediction equations for determining FFM, using DXA and ADP as the two reference methods.

2. Develop BIA prediction equations for determining SMM_a, with DXA as the reference method.

3. Construct %FM and SMM_a reference centiles.

4. Assess BC differences between the SA study population and available data for WE children and adolescents.

5. Construct WC reference centiles with cut-offs in line with the SA adult-cut offs adopted in India.

6. Develop a portable tool for measuring SH for the determination of RLL.

7. To compare RLL of the study population with available WE data.

CHAPTER 3: General Methods

3.1 Introduction

This chapter provides an overview of the standard operating procedures (SOP) used for all measurements taken in the individual studies described in the relevant chapters, the recruitment procedures followed, and some of the common statistical procedures used. This chapter is divided into three sections: (i) anthropometric measures, which define the methods used for measuring body size and proportions; (ii) body composition measures to define the methods used in body composition analysis such as % fat mass (FM) and skeletal muscle mass (SMM) or fat-free mass (FFM) using bioelectrical impedance analysis (BIA), dual-energy x-ray absorptiometry (DXA) and air displacement plethysmography (ADP); and (iii) common statistical procedures used such as the LMS method (Cole & Green, 1992), which was used to construct centile curves for %FM and appendicular SMM (SMMa) (Chapter 5), and waist circumference (WC) (Chapter 7), and the methodology used for determining the reliability of all anthropometric measures. The importance of using SOPs is to ensure that the data was collected with precision, to limit discrepancies within and between studies, and to enable the study to be effectively replicated, without compromising its validity (Bellisari and Roche, 2005).

3.2 Recruitment Procedures

An opportunistic sample of healthy children and adolescents (based on self-report) was recruited for the studies conducted in this thesis. Data from the field-based studies were added to an existing data set of body composition and anthropometric measures of a large South Asian (SA) cohort of 1,459 UK school-aged children from a previous study conducted at this university that were recruited from primary and secondary schools within London and Greater London (Radia, 2010), although no leg length data was available in that study. The majority of SA subjects from the external mixed ethnicity cohort were from Tower Hamlets (~33% Bangladeshi), Newham (~10% Bangladeshi), Harrow (~22% Indian), Ealing and Hounslow (~17%). As one of the aims of the field-based studies was to make body composition and anthropometric comparisons within the SA ethnic group, and the external data

set lacked sufficient numbers from the Pakistani sub-group, local mosques and Muslim community centres in London boroughs were approached in an attempt to capture a large number of Pakistanis in the sample. However, this proved difficult as the majority of subjects were of Indian descent. This was not considered to be a major setback as Pakistan was only formed in 1947; prior to that it was part of India, thus all SA participants would be originally of Indian descent and it was assumed that differences in body composition within SA ethnicities was more likely to be related to lifestyle rather than genetic factors.

The target age range was 5-19y to mirror the most recent WHO growth reference standards (de Onis et al., 2007), although the cut-off was 23y in accordance with the UK90 reference charts for BMI (Cole et al., 1995). Efforts were made to restrict the age range, and preferentially recruit the majority of subjects in the 6-18y age range to ensure a wide variation in body compositions, as the major changes to body composition occur during later childhood and adolescence, with adiposity rebound occurring at approximately age 6y (Cole et al., 1995).

Ethical approval was obtained from London Metropolitan University's Ethics Committee before subject recruitment (Appendix B1). Flyers were initially distributed with reply slips collected at stands held at community events (Appendix B2). Consent forms, letters of invitation, and information packs (Appendix B3-B5) were distributed to interested participants, with signed consent required by subjects ≥16y and signed parental consent, together with child assent, required for children <16y. Subjects were also requested to complete a brief questionnaire on gender, self-assigned ethnicity, and date of birth, prior to conducting any of the tests.

3.3 Anthropometric Measures – Accuracy & Reliability

Anthropometry is the measurement of the body including its size (e.g. body weight and stature) and proportion (e.g. BMI, or sitting height ratio (SHR); Heyward & Wagner, 2004). To minimise measurement error, it is important to follow standardised procedures, as measurement error will affect the accuracy and reliability of the results (Heyward & Wagner, 2004; Ulijaszek & Kerr, 1999). Accuracy of measurement refers to how 'true' the value of the measure is (Mueller

& Martorell, 1988), and poor accuracy may be the result of equipment error, issues with the client, or observer error due to inexperience of the technician (Heyward & Wagner, 2004; Ulijaszek & Kerr, 1999). Thus to minimise inaccuracy, a single observer took all anthropometric measures for the studies conducted in this thesis and followed standardised procedures including regular calibration and checking of equipment, and practised taking all anthropometric measures under supervision prior to data collection; where secondary data was used the researcher ensured that the same measurement protocol was followed.

Reliability refers to precision of the results, or whether repeated measures would give a similar result, and is affected by intra- (one observer) or inter- (more than one observer) differences (Ulijaszek & Kerr, 1999). In addition to measuring body size and proportionality, anthropometric measures are also used as proxy markers of adiposity, such as BMI (weight (Wt; kg)/ height (Ht; m^2), and WC respectively. These measures are also used as part of the more complex body composition assessment methods, such as BIA (BIA), and DXA. To assess reliability of the measurement method, certain statistical procedures are recommended, which are explained below in section 3.4 on common statistical procedures.

3.3.1 Stature

Stature or standing Ht was measured to the nearest 0.1cm using a portable stadiometer (Leicester Height Measure (LHM); Figure 3.1). Participants were asked to remove their shoes and stand as upright as possible on the base plate, with feet positioned on the foot markers, and arms hanging loosely either side of the body, with palms facing inwards, and shoulders relaxed.

The child was requested to hold their head erect with eyes facing directly forward. From the left side of the participant, the head was positioned by the researcher, in the Frankfort Plane position, whereby an imaginary horizontal line was drawn from the upper margin of the external auditory meatus and the lower border of the eye orbit (Martin et al., 1988). The horizontal top-plate of the stadiometer was then lowered to sit on the head compressing any hair, and the Ht reading was taken from the red arrow pointer within the measuring window. The LHM was regularly calibrated using a 1m rule, and each Ht measurement was taken once.

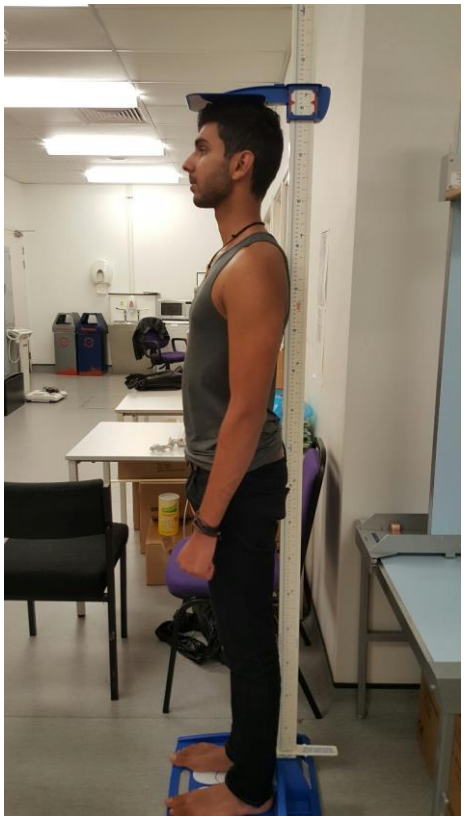


Figure 3.1 Leicester Height Measure with participant

3.3.2 Body Weight

Body weight was measured to the nearest 0.1kg, in light indoor clothing with bare feet, on the Tanita BC418-MA (Tokyo, Japan) Bioelectrical Impedance Analyser (BIA; see Figure 3.2 below), which incorporates an electronic digital scale. A correction factor of 0.5kg was applied to account for clothing. Prior to measuring, the children were requested to remove any heavy items of clothing and jewellery, as well as mobile phones. The equipment was regularly calibrated and serviced according to the manufacturer's guidelines.

3.3.3 Waist Circumference

Waist circumference was measured using a flexible tape to the nearest 0.1cm above a single layer of clothing, typically a t-shirt, vest, or school shirt, positioned midway between the top of the iliac crest and lower border of the bottom rib at the narrowest part of the torso, following a standardised procedure (Heyward & Wagner, 2004), with 0.5cm deducted from the measurement to account for clothing (McCarthy et al., 2003). In the standing position, subjects were asked to raise their arms horizontal to the floor, whilst the tape measure was passed around the body,

and lower the arms during actual measurement, which was taken at the end of normal expiration.

3.3.4 Sitting Height and Leg Length

Following the successful development of a field-based sitting height measure (SHM; see chapter 6) sitting height (SH) was measured using the adapted Leicester Height Measure. All measures were taken from the left side of the body following standard guidelines (Cameron et al., 1981). Subjects were requested to sit onto the SHM which was placed on a standard table, with the back of the knees aligned with the table edge, so that the legs could hang freely directly at the knee joint. During this process, any shift in the SHM was corrected by requesting the participant to rise up slightly, using their hands for support, to enable the researcher to make appropriate adjustments.

Subjects were requested to sit as upright as possible, with shoulders relaxed and hands placed on the thighs. To prevent slumping, the researcher placed one hand at the base of the spine and the other at the top of the shoulder pressing it slightly backwards, prior to adjusting the head into the Frankfort Plane position as described above for measuring Ht (section 2.2.1). The sliding top plate was then lowered and positioned directly onto the head, compressing any hair. SH was recorded using the same methodology as described for measuring stature (section 2.1.1), with 2cm deducted from the reading to account for the Ht of the removable base (see chapter 6). The SHM was regularly calibrated using a 1m rule, and for the field-based study each measurement was taken once. Leg length (LL) was calculated by deducting SH from the stature measurement.

3.4 Body Composition Measures

The BIA validation study (chapter 3), and the field study (chapter 4), used the BC-418MA (Tanita, Tokyo) analyser. This BIA model was validated using 2 reference methods: DXA (Norland XR-8000) pencil beam scanner, and ADP. General methods used for all 3 technologies are described here. All measures were conducted in a single session for each subject, to minimise variation.

3.4.1 Bioelectrical Impedance Analysis

The analyser was placed on the floor on a stable flat surface, and all measures were conducted indoors at normal room temperature. Subjects were requested to empty their bladders prior to BIA testing to prevent any bias in the results. Measurements were conducted in light indoor clothing, with bare feet. Prior to subjects standing on the analyser, age, sex, and Ht were entered into the analyser (section 2.2.1). All subjects were entered as 'standard' body type, as none of the subjects recruited were considered to have an athletic build. Wt was measured on the BIA as detailed above (section 2.2.2). The same analyser was used for all subjects in both the validation and field studies.

Correct foot placement was explained to participants to ensure that the whole foot was in contact with the electrodes on the metal foot-plates, with any clothing raised to ankle Ht to avoid being trapped under foot. Subjects were then requested to grasp each of the handles (Figure 3.2), and hold the handles either side of the body, away from the legs at an approximate 45° angle.

Figure 3.2 Tanita BC418-MA BIA image with participant

3.4.2 Dual-Energy X-ray Absorptiometry

Prior to measurement, the scanner was calibrated using a phantom provided by the manufacturer. Subjects were measured in light indoor clothing, without shoes, and were requested to remove any metal on their clothing or bodies such as belts, watches, jewellery, and mobile phones, prior to testing. Subjects were carefully positioned lying in the supine position on the scanning table, with arms and legs placed within the scanning perimeter as marked out on the table, with palms facing down (Figure 3.3). Where possible, arms were positioned slightly away from the body, and legs were positioned slightly apart, for improved scan clarity.

During scanning, which takes approximately 5 minutes, subjects were requested to lie as still as possible and close their eyes, with the head turned to the right. The importance of not looking at the laser during operation was clearly explained to participants. The DXA was linked up to a computer, and scanning software (Norland-Illuminatus) was used to analyse the pixelated image, which provided output for % whole body fat (including Siri and Brozek %fat), as well as regional FM and LM (g; see chapter 4 Figure 4.2).

Figure 3.3 Norland XR-800 DXA scanner with participant & computer software images

3.4.3 Air Displacement Plethysmography

In this study, the ADP system employed was the Bod Pod® Body Composition System (Life Measurement Instruments, Concord, CA; Figure 3.4), as described in chapter 2 (section 2.5.3). Prior to subject measurements, the Bod Pod was calibrated using a 50l cylinder, following the on-screen step-by-step procedures. Repeat calibrations were conducted between subjects where possible. Subjects were measured in skin-tight bathing suits, with long sleeves and skin-tight leggings available if requested, and a swim-cap was used to cover the hair to minimise measurement error due to extra volume created by loose clothing and hair (McCrory et al., 1995). Predicted rather than measured lung volume was used. A male and female researcher were available to measure participants, if a same-sex researcher was requested. The test took 3-5 minutes, and the participant was first weighed to the nearest 100g on a set of electronic scales linked up to the Bod Pod computer before entering the chamber.

Figure 3.4 Bod Pod® images with participant in-situ

3.5 Data Handling & Statistical Procedures

To ensure anonymity, all participant data entered into Excel (Microsoft® Excel® for Mac 2011 version 14.4.8) and SPSS (IBM® SPSS® version 22 for Mac) was appropriately coded using a system of letters and numbers at time of data collection, recording date of measurement, date of birth, gender, and all anthropometric measures. Secondary SA (ASD) and WELI anthropometric data from a previous study conducted at this university was used for the field-based studies (Chapters 5

& 7) to maximise the SA sample size and also to provide data for comparisons with a contemporary WELI population.

3.5.1 Determination of Anthropometric Measurement Error – reliability

As stated in section 3.2, precision of measurement in anthropometry, is an assessment of reliability of results, with reference to repeated measures of a sample population taken by a single or several observers giving equivalent or very close to equivalent results. The technical error of measurement (TEM) is a commonly used method for providing an indication of the level of intra- or inter-observer precision in anthropometric measures (Mueller & Martorell, 1988). TEM is a measure of standard deviation, which is used to calculate the proportion of the total standard deviation of the sample population that results from measurement error. TEM is determined by taking repeated measurements of each subject either by one or more observers, where the difference between the measurements is entered into an appropriate equation. When only two measurements for each subject are taken, TEM is determined using the following equation:

(1) $$TEM = \sqrt{(\sum D^2/2N}$$

Where D = the difference between the two measurements, and N = the number of subjects measured.

When more than two measurements of each subject are taken, a more complex equation is used:

(2) $$TEM = \sqrt{((\sum_1^N((\sum_1^K M^2) - ((\sum_1^K M)^2/K)))/N(K-1))}$$

Where N = number of subjects; K = the number of measurements, and M = the actual measurement.

AS TEM has the same unit value as the unit of measurement, and large measurement values result in larger TEM values, comparisons between different measurements is difficult (Ulijaszek & Kerr, 1999).

To overcome this issue, use of relative (%) is recommended, which converts TEM to a coefficient of variation value (CV; Norton & Olds, 1996 in Ulijaszek & Kerr, 1999) using the following equation:

(3) %TEM = (TEM/mean) x 100

Where the mean value is the mean of the measured variable. However, even %TEM is problematic when making comparisons between different measures as it is negatively associated with the size of the measure, i.e. for the same TEM, large mean measurement values result in comparatively smaller %TEM values than small mean values (Roebuck et al., 1975). Due to these limitations, the coefficient of reliability (R) is considered more suitable (Mueller & Martorell, 1988). This measure ranges from 0 to 1, and is determined using the following equation:

(4) $R = 1 - (\text{total } TEM^2/SD^2)$

The value of R represents the proportion of total variance between all measured variables that is free of measurement error, thus the closer the value of R is to 1 indicates the smaller the margin of error (Mueller & Martorell, 1988). This method enables comparisons between different anthropometric measures to be made, as the absolute size of the measure does not impact the value of R (Mueller & Martorell, 1988).

To determine anthropometric measurement error for Ht, waist circumference (WC), and SH a sub-sample of 10 subjects was measured 5 times. Table 3.1 provides results for the TEM (based on equation 2), %TEM (equation 3), and R (equation 4). Different subjects were measured repeatedly for the three types of measurements taken to avoid fatigue. The results in Table 3.1 illustrate the problems with TEM, where absolute (cm) TEM is the greatest for height and smallest for relative (%) TEM, whilst the R-value is the same for all three measurements. All R-values were close to 1, indicating a low intra-observer margin of error.

Table 3.1: Anthropometric measures for Ht, WC, & SH measures (mean ± sd) for 10 subjects together with absolute (cm) & relative (%) TEM, and R- value

Subj. #	1	2	3	4	5	6	7	8	9	10	TEM (cm)	TEM (%)	R
Ht (cm)	180.9 ±0.11	155.5 ±0.32	168.5 ±0.29	185.5 ±0.16	149.2 ±0.69	172.1 ±0.15	154.4 ±0.26	182.5 ±0.32	165.6 ±0.29	187.3 ±0.19	0.33	0.19	0.99
WC (cm)	77.3 ±0.24	68.5 ±0.13	84.4 ±0.24	59.6 ±0.19	73.5 ±0.26	92.3 ±0.24	103.8 ±0.19	58.6 ±0.19	87.5 ±0.19	102.5 ±0.17	0.24	0.30	0.99
SH (cm)	94.1 ±0.16	84.6 ±0.12	75.7 ±0.21	95.6 ±0.15	69.3 ±0.24	91.2 ±0.22	83.4 ±0.24	95.5 ±0.26	73.5 ±0.24	98.4 ±0.21	0.21	0.25	0.99

Ht = Height; WC = Waist circumference; SH = Sitting height; TEM = technical error of measurement; R = reliability (see section 3.5.1)

3.5.2 Standard Deviation Scores and the LMS method

Sex-specific standard deviation scores (SDS) or Z-scores were generated in Excel, for all anthropometric variables including Ht (cm), Wt (kg), BMI, WC (cm), SH and LL, and also %FM, using the downloadable Microsoft Excel add-in LMSgrowth (www.healthforallchildren.com) which contains the British 1990 growth reference data (Cole et al., 1995). Presenting data in SDS format against British 1990 reference data enables population comparisons to be made against the relevant measures with adjustment made for changes due to age (Cole & Green, 1992).

Sex-specific smoothed percentile curves for WC, %FM and SMMa were constructed by importing data from Microsoft Excel into the downloadable LMS Chartmaker Light software (www.healthforallchildren.com). The LMS method (Cole & Green, 1992), accounts for skewness, and normalises the data for each age group based on three parameters: The Box-Cox power (Lambda 'λ'), the mean (Mu 'μ'), and the coefficient of variation (Sigma 'σ'), where the term 'LMS' is derived from the initials of each parameter. The main assumption of the LMS method is that following appropriate power transformation the data will be normally distributed, and whilst this method does not account for kurtosis, this is considered less important than skewness when adjusting for normality (Cole & Green, 1992). Exact SDS for each individual can be converted to a centile value using normal distribution tables (Cole et al., 1995).

3.5.3 Exploration of Data

Prior to conducting any parametric statistical tests in SPSS, the descriptive statistics of SDS for Ht, Wt, BMI, WC, and %FM were explored to check for any outliers or extreme cases (boxplots) and for normality of distribution by assessing histograms, mean and 5% trimmed mean, skewness and kurtosis, and the test of normality based on the Kolmogorov-Smirnov output, following recommended procedures (Field, 2012; Pallant, 2013). Selected SPPS outputs showing normality of distribution are presented in Appendix C. For the BIA validation study (Chapter 4), any extreme cases or outliers that exerted an undue influence on the mean were removed from the dataset. To assess whether the cohort for the BIA validation study were representative of the wider SA population, sex-specific comparisons with the large external SA dataset for all measured SDS were made (Chapter 4, section 4.5). Examination of histograms indicated that for the validation study the data was normally distributed, which was confirmed by a non-significant (Sig.value >0.05) Kolmogorov-Smirnov statistic. SDS for %FM had two missing values for boys and one for girls, as the UK90 data was limited to age 20y for this variable. WC data also had missing data, as the UK90 data for this variable was limited to age ≤ 16y.

The normality tests (Appendix C) for the external datasets for the SA ASD boys, and the WELI boys and girls revealed some of the anthropometric SDS measures violated the assumptions of normality with a significant (≤0.05) Kolmogorov-Smirnov statistic. However, according to the 'central limit theorem' (Field, 2012 p.170-172) in large sample sizes normality can be assumed regardless of the shape of the sample data, and should not affect significance tests. Therefore, only parametric statistics tests were conducted in each of the studies. Normality tests for Ethnic Differences in Leg length (Chapter 8) are presented in Appendix D. All measured variables in this study were normally distributed, confirmed by a non-significant (Sig.value >0.05) Kolmogorov-Smirnov statistic.

CHAPTER 4: Validation Study of Tanita Bioelectrical Impedance Scales using DXA and ADP

4.1 Introduction

The BC-418 (Tanita, Tokyo) is a widely available bioelectrical impedance analyser (BIA) for clinical use, using 8-contact electrodes to measure electrical impedance (Z) in the standing position, in all four limbs. It provides whole body and segmental estimates for fat mass (FM), fat-free mass (FFM), and segmental predicted muscle mass (PMM) for all four limbs, as well as the trunk and head (Pietrobelli et al., 2004; McCarthy et al., 2006). The system uses inbuilt equations obtained from regression analyses, with DXA as the reference method (Pietrobelli et al., 2004). This model has also been validated in a mixed paediatric population of predominantly white European (WE) ethnicity, against dual-energy x-ray absorptiometry (DXA) and air displacement plethysmography (ADP) (BodPod®; Pietrobelli et al., 2005), which are considered valid, reliable and suitable methods for determining body composition (BC) in children and adolescents (Fuller et al., 2002; Heymsfield et al., 1990; Lohman et al., 2006).

Essentially, BIA, based on the 2-C model (where body mass = FM + FFM), is based on the principle that height (Ht) squared divided by impedance (Ht^2/Z), is proportional to the volume of total body water (TBW; Haroun et al., 2010; Kyle et al., 2004). TBW or FFM is indirectly estimated by regression analysis between the BIA output Ht^2/Z and a criterion method such as hydrometry or DXA respectively, or use of a 4-C model, where FM and FFM are determined (see Chapter 2 for detailed review). As FM is anhydrous, FFM can be estimated from TBW, where the hydration of FFM is considered constant (Heyward & Wagner, 2004; Kyle et al., 2004). For example, if the hydration of FFM was assumed to be 73%, then FFM could be determined by dividing TBW by 73% (i.e. FFM = TBW/0.73). However, due to population differences in the hydration of FFM, particularly between ethnic groups, genders, and across childhood and adolescence, population specific prediction equations of FFM or TBW are recommended (Baumgartner et al., 1991; Ellis, 2000; Wang et al., 2005).

The BIA in-built equations for the BC-418 model were validated using a WE sample population (Pietrobelli et al., 2004), and the use of such equations are not necessarily recommended in other ethnic groups, due to ethnic differences in the hydration of FFM (Frisard et al., 2005; Jebb et al., 2000; Sun et al., 2003; Kyle et al., 2004). Manufacturers do not provide details of how these equations are formulated, as this is considered proprietary information. Many centres have developed paediatric FFM prediction equations using BIA (Kyle et al., 2004; Haroun et al., 2010; Sluyter et al., 2010; Williams et al., 2007), which are known to vary not only due to ethnic differences in hydration of FFM (Chumlea & Sun, 2005), but also by reference method used, including variations between and within DXA models (Helba & Binkovitz, 2009; Plank, 2005).

Very few studies have developed FFM prediction equations for SA children and adolescents, that in general show that compared to reference methods, BIA underestimates FM in the South Asian (SA) population (Haroun et al., 2010; Khan et al., 2012; Sluyter et al., 2010). Only one study (Sluyter et al., 2010) was identified that developed a BIA prediction equation using the BC-418 BIA model, which was validated using a DXA pencil-beam scanner (GE Lunar, Madison, WI) as the reference method. However, this study was limited to Asian adolescents (aged 12-19y) from several different countries, and whilst the majority of the cohort were Indian, some participants were from other Asian countries including Cambodia and China, which may have impacted the predictive quality of the equation (see chapter 2 section 2.3.1.6, for more detail). In that study, compared to DXA, BIA on average significantly ($P<0.001$) underestimated %FM by 2.84%. There was also a tendency to overestimate %FM in lean individuals, and underestimate %FM in individuals with high adiposity levels.

A validation study has compared ethnic differences in TBW, FM and FFM using in-built equations, between SA, WE, and Black children aged 11-15y using a bipolar BIA instrument (Tanita TBF-300) with D_2O as the reference method; for TBW no significant bias was observed in the white population, and SA female population, however, TBW was significantly overestimated in the male SA population, and underestimated in the black population (Haroun et al., 2010). FM was also

underestimated in all ethnic groups apart from black females, with the greatest underestimation occurring in the SA population. Variation in body geometry and distribution of FM and FFM between ethnic groups, confounded by the use of a bipolar BIA model were considered possible sources of error in that study (Haroun et al., 2010). Similar to the finding by Sluyter et al. (2010), BIA tended to underestimate TBW (thus overestimate FM) in individuals with higher levels of TBW (i.e. lean subjects), and TBW was overestimated (thus underestimating FM) in individuals with lower levels of TBW (i.e. subjects with greater adiposity).

An advantage of the BC-418 analyser is that in addition to estimating total body FM and FFM, it also provides an estimation of appendicular fat and skeletal muscle mass (FM_a; SMM_a), with evidence of a strong correlation between impedance and SMM_a (Haroun et al., 2010; Pietrobelli et al., 1998). As there is an established link between the distribution of FM and SMM, with abdominal adiposity and lower SMM_a considered to be greater risk factors for type 2 diabetes mellitus (T2DM) and cardiovascular disease (CVD), data on SMM_a could provide health practitioners with further information on the additional health risks associated with low SMM_a.

The BC-418 BIA has the potential of providing a simple and reliable means of determining body composition within the SA child and adolescent population, if suitable prediction equations were available for this ethnic group. References similar to those developed for WE children and adolescents for %FM (McCarthy et al., 2006) and % SMM_a (McCarthy et al., 2014), could be used in conjunction with existing BMI and WC references, which would overcome the limitations of relying on BMI alone to identify health risks, particularly in this population group, where BMI has been shown to be particularly unsuitable (see Chapter 2 – section 2.3).

4.1.1 Study Aims

In view of the limitations of the current BC418 equations for the SA child and adolescent population, the aims of this study were to:

i) Validate the use of the BC418-MA (Tanita, Tokyo) BIA for SA children and adolescents in the UK (aged 5-20y), by developing sex-specific prediction equations for FFM, using DXA and ADP as the reference methods, from which %FM can be determined.

ii) Validate the use of BIA (BC418-MA) with DXA to determine $\%SMM_a$ for SA children and adolescents in the UK (aged 5-20y).

iii) Evaluate the SA FFM prediction equations produced by Sluyter et al. (2010), and Haroun et al. (2010), on the sample population as well as on the large SA cohort, within the respective age ranges.

4.2 Study Design

Body composition (BC) analysis was conducted using the tetrapolar Tanita BC-418MA Segmental Body Composition Analyser (Tanita Corporation, Tokyo, Japan). To ensure validity of the Tanita analyser for SA children, the equipment was validated against a DXA scanner and an ADP analyser in a SA paediatric population, with the development of ethnic-specific prediction equations for FFM. Validation tests were carried out in line with previous methods (Haroun et al., 2010; McCarthy et al., 2006; Pietrobelli et al., 2004; Sluyter et al., 2010).

Previous validation studies have used a sample size of between 30-50 subjects, with equal numbers from each sex (Haroun et al., 2010; Lazzer et al., 2003; Pietrobelli et al., 2004). However, according to Haroun and colleagues (2010) more important than sample size per se, is ensuring that the distribution of body composition within the sample has wide variability, to enhance the predictive accuracy of the results against the larger population. Comparisons between the sample population and a large (n= 1,459) existing data set of SA UK school-aged children (see General Methods chapter, section 3.2) were also made, to ensure that the sample was representative of the wider SA population. In addition, comparisons were also made between SA and WE variables converted to SDS format based on the UK90 reference

data (Cole et al., 1995), to assess differences between groups (see General Methods chapter section 3.4.2).

4.3 Subject Recruitment & Experimental Procedures

Healthy children and adolescents (based on self-report) were recruited for this study, with a target of 50 participants, divided equally by sex. Local mosques and Muslim community centres in North London were approached, following recruitment procedures described in Chapter 3 (General Methods, section 3.2).

4.3.1 Anthropometric Measures

Anthropometric measures including Ht, weight (Wt), and waist circumference (WC) were conducted following standardised protocols, as detailed in Chapter 3 (General Methods, section 3.3).

4.3.2 Bioelectrical Impedance Analysis

The BC-418 has 8 stainless steel contact electrodes; 2 on each footplate and 2 in each hand grip (Haroun et al., 2010; Figure 4.1). A pre-set imperceptible electrical signal is passed through the subject's tissues and the fall in impedance is used to estimate total body water (TBW), from which FFM and FM is estimated. Detailed procedures for measuring subjects are explained in Chapter 3 (General Methods). Data for FFM and %FM for the whole body, and SMM for all four limbs was recorded.

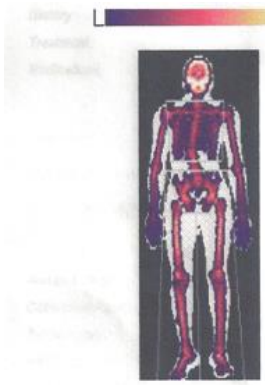
Figure 4.1 Tanita BC418-MA BIA

4.3.3 DXA

The Norland XR-800 pencil beam scanner was used to measure %total body fat, %Siri body fat, lean tissue mass (LTM) and appendicular (i.e. all four limbs) lean tissue mass (ALTM) or SMM_a. Chapter 3 (General Methods) provides standard operating procedures and the principles of DXA are explained in the Literature Review (Chapter 2 section 2.5.2). The scanner is linked to software, which provides whole body and regional (head, trunk, and appendicular) compositional estimates for FM, FFM, LTM, and bone mineral mass along with a graphical image (Figure 4.2).

The Norland XR-800 scanner provides absolute values for bone mineral (g/cm^2), soft tissue FFM (g), and FM (g). %FM values are also provided for: total fat; Siri UWE (Underwater Equation) fat; Brozek UWE fat; and soft tissue fat (Figure 4.2). The Siri UWE %FM is used as the criterion value for determining where the individual lies in terms of adiposity, in comparison to national references. The method for determining this value is not provided by the manufacturers, and is considered proprietary information.

As a criterion measure, DXA is considered accurate for body composition proportionality i.e. %FM and %FFM. However, DXA Wt is derived from the sum of 3 component parts, i.e. bone-mineral content (BMC), soft-tissue FFM (described as LM), and FM (Figure 4.2), and some studies have reported discrepancies between DXA derived Wt and scale Wt (Friedl et al., 1994; Korth et al., 2007; Nelson et al., 1996; Rush et al., 2009).



Bone image not for diagnosis.

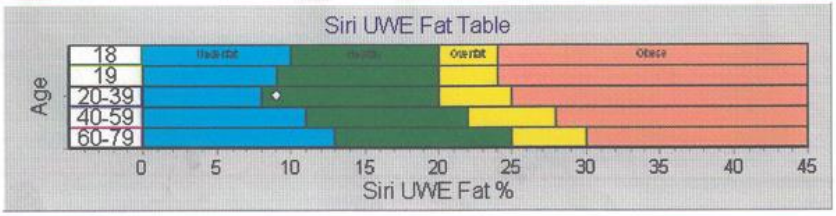
Whole Body on: 8/30/2011 4:15:50 PM

Region	%
Total Fat	12
Siri UWE Fat	9
Brozek UWE Fat	9
Soft Tissue Fat	13
TBMC/FFM	5

Region	BMD (g/cm²)	BMC (g)	Area (cm²)	Length (cm)	Width (cm)	Lean Mass (g)	Fat Mass (g)	T-Score	Young Ref. (%)	Z-Score	Age Match (%)	ST Change (%)	ST Change (%/yr)	LT Change (%)	LT Change (%/yr)
Total	0.8040	2166	2695			41845	5983	*****	*****	*****	*****	*****	*****	*****	*****
Head	1.719	414.1	240.8			2873	648.4	*****	*****	*****	*****	*****	*****	*****	*****
Chest	0.6961	376.4	540.7			10018	1244	*****	*****	*****	*****	*****	*****	*****	*****

STD CV for Total BMC: 0.9 See Guide for other CVs.
6.5 mm X 13.0 mm, 260 mm/s, 66.95 cm, Rev. 4.3.1/2.3.1 Calib. 08/30/11 S/N 8580

Comments:



Scan Information

Scan Type:	Whole Body	***Resolution:***	13.0 x 6.5
Analysis Date:	11/9/2011 11:47:21 AM	***Speed:***	260 mm/s
Calibration Date:	08/30/11	***Width:***	67.0
Technologist:		***Host/Scanner:***	4.3.1/2.3.1
Physician:		***Analysis/Revision:***	4.3.1

Detailed Results

Region	BMD (g/cm²)	BMC (g)	Area (cm²)	Length (cm)	Width (cm)	Lean Mass (g)	Fat Mass (g)	T-Score	Young Ref. (%)	Z-Score	Age Match (%)	ST Change (%)	ST Change (%/yr)	LT Change (%)	LT Change (%/yr)
Midriff	0.9272	34.01	36.68			1818	335.6	*****	*****	*****	*****	*****	*****	*****	*****
Pelvis	0.7626	224.0	293.7			7490	226.0	*****	*****	*****	*****	*****	*****	*****	*****
Left Leg	0.8820	405.1	459.3			7358	1281	*****	*****	*****	*****	*****	*****	*****	*****
Right Leg	0.8497	395.2	465.1			7097	1473	*****	*****	*****	*****	*****	*****	*****	*****
Left Arm	0.4841	165.9	342.7			2731	409.7	*****	*****	*****	*****	*****	*****	*****	*****
Right Arm	0.4808	151.8	315.6			2459	366.5	*****	*****	*****	*****	*****	*****	*****	*****

Figure 4.2 Example of DXA output of whole-body scan (from Norland XR-800 pencil beam scanner). DXA = dual-energy x-ray absorptiometry

4.3.4 ADP

ADP (Bod Pod®) is a 2-C method, which measures body volume and mass, from which body density (Db) is determined (McCrory et al., 1995). %FM is determined using the Siri (1961) equation: %FM = [(4.95/Db - 4.50) x 100)], and FFM is determined by subtracting FM from body Wt. Chapter 3 General Methods, provides detailed operating procedures and the principles of ADP are explained in the Literature Review (Chapter 2 section 2.5.3).

4.4 Statistical Analyses

Preliminary analyses of the data were conducted to assess for normal distribution (See Chapter 3 General Methods section 3.4.3). Data for Ht, Wt, BMI, WC, and %FM were also converted to standard deviation scores (SDS) or Z scores, using LMS Growth (Cole et al., 1995) software, to compare with UK90 reference data.

For validation of the BC418 analyser, the relationship and difference between BIA and the reference methods DXA and ADP (BodPod), for absolute and relative FM and FFM (kg and %), were compared, firstly for the whole sample and then split by gender. Additionally, to assess whether the sample was representative of the wider population, comparisons were made with the external group, as explained in section 4.2 above. Statistical analyses involved paired sample correlations, t-tests, and ANOVA, as well as simple multiple linear regression, using the Statistical Package for Social Sciences (SPSS v.20 for Mac; IBM.com). Significant values were computed at the $P<0.05$ level unless otherwise stated. Segmental comparisons between BIA and DXA also included trunk and appendicular body composition measures. Bland-Altman (Bland & Altman, 1986) analyses were also conducted for between-method bias for all measured variables. New equations were developed for predicting FM and FFM following regression with BIA and the reference method.

4.5 Results

A total of 53 (25 females and 28 males) healthy children and adolescents aged 5-21y, were recruited for the study. Initial analyses of the sample data set, revealed one extreme case and one outlier for Wt and FM within the female sample, which when compared with the external data set was exerting an undue influence on the

whole sample mean SDS; these two cases were therefore removed from the analyses. The female participant presenting as an extreme case in the data analysis did not fit within the DXA scanning range due to her large size, and one male participant had to be excluded from the analyses, as he was discovered to have a mobile phone in his pocket, after the scan had been completed. Of the final sample of 50 subjects (23 females and 27 males), 10 were Bangladeshi and 2 were Pakistani, therefore within-group comparisons were not made. 3 (2 females and 1 male) declined to enter the BodPod resulting in missing data for this measure.

4.5.1 Subject Characteristics

The subject characteristics of the final sample are summarised in Table 4.1, which also includes data from the external group (see section 4.2 above). 121 (boys n = 82 and girls n = 39) subjects were recruited for the field-based study, and data taken from the previous body composition study of 1459 (boys n = 584 and girls n = 875) school-aged children was added. This external dataset was included to assess whether the sample was representative of the wider population, as explained previously, and to apply any correction factors developed in this study.

Compared to UK90 reference data, the study group sample as a whole, was on average lighter (-0.29 SDS), shorter (-0.30 SDS), had a lower BMI (-0.29 SDS), but a higher %FM (0.23 SDS; based on BIA), and WC (0.35 SDS). For boys, on average, the sample was lighter (-0.30 SDS), slightly shorter (-0.07 SDS), with a lower BMI (-0.48 SDS), and a higher %body fat (0.35 SDS), and slightly higher WC (0.01 SDS). Similarly, the female sample was on average lighter (-0.29 SDS), shorter (-0.57 SDS), with a slightly lower mean BMI (-0.07 SDS), and a higher %FM (0.10 SDS), and WC (0.75 SDS). There were no statistically significant differences between the sexes for most of the measured variables in Table 4.1, except for BIA FM (kg; P= 0.008), and BIA %FM (P< 0.001). Similarly, the SA external group, for the sample as a whole, and for boys and girls respectively, compared to the UK90 reference data, was on average lighter (-0.11; -0.06; -0.16 SDS), very slightly shorter (-0.06; -0.03; -0.08 SDS); with a lower BMI (-0.17; -0.11; -0.21 SDS), and a higher %FM (0.37; 0.53; 0.25 SDS), and WC (0.25; 0.31; 0.20 SDS).

Same sex comparisons were also made between the study group and the SA external group for all measured variables (Table 4.1). The mean age of both the male (12.27y) and female (13.5y) sample of the study group was significantly greater ($P< 0.05$) than that of the external group (mean age = 9.63y & 9.12y respectively). Likewise, there were significant differences between most of the absolute values of the measured variables at the $P< 0.05$ level. However, for SDS values, apart from a significant ($P< 0.05$) difference in Ht for the female sample only (Ht SDS = -0.57; -0.08 for the study group and external group respectively), no significant differences were observed between the SDS values for any of the other SDS measured variables.

Table 4.1: Subject characteristics (mean ±sd) and comparisons of mean values between boys and girls of study group, and same sex comparisons between BIA validation study group and SA external group.

Variables	BIA Validation Study Group			SA External Group		
	All (n=50)	**Boys (n=27)**	**Girls (n=23)**	**All (n=1580)**	**Boys (n=666)**	**Girls (n=914)**
Decimal Age (years) (Range) (y)	12.84 ± 4.26 (5-21)	12.27 ± 4.25 (5-21)	13.5 ± 4.27 (5-21)	9.63 ± 2.71 (4-21)	9.12* ± 2.74 (4-21)	10.00* ± 2.63 (4-21)
Wt (kg)	42.35 ± 17.19	41.54 ± 19.46	43.31 ± 14.45	33.35 ± 13.60	31.69* ± 13.54	34.56* ± 13.51
Wt SDS	-0.29 ± 1.04	-0.30 ± 1.15	-0.29 ± 0.92	-0.11 ± 1.34	-0.06 ± 1.37	-0.16 ± 1.32
Ht (m)	1.48 ± 0.18	1.49 ± 0.21	1.47 ± 0.13	1.36 ± 0.17	1.33* ± 0.18	1.38* ± 0.17
Ht SDS	-0.30 ± 1.11	-0.07 ± 0.91	-0.57 ± 1.28	-0.06 ± 1.01	-0.03 ± 1.00	-0.08* ± 1.01
BMI (kg/m^2)	18.5 ± 4.31	17.7 ± 4.05	19.49 ± 4.50	17.3 ± 3.70	17.0 ± 3.56	17.51* ± 3.79
BMI SDS	-0.29 ± 1.37	-0.48 ± 1.37	-0.07 ± 1.36	-0.17 ± 1.46	-0.11 ± 1.52	-0.21 ± 1.42
WC (cm) (n=48)	66.06 ± 11.57	65.36 ± 12.26	66.89 ± 10.92	59.99 ± 10.07	60.34* ± 10.56	59.73* ± 9.70
WC SDS (n=39)	0.35 ± 1.29	0.01 ± 1.12	0.75 ± 1.38	0.25 ± 1.41	0.31± 1.36	0.20 ± 1.45
BIA FM (kg)	9.38 ± 5.32	7.36* ± 3.78	11.38*± 5.91	8.06 ± 5.43	6.92 ± 4.98	8.89* ± 5.60
BIA %FM	21.32 ± 7.08	18.36** ± 4.78	24.79** ± 5.91	22.49 ± 6.16	20.37 ± 6.09	24.03 ± 5.75
BIA FFM (kg)	32.30 ± 12.49	33.86 ± 15.96	31.93 ± 9.29	25.30 ± 8.92	24.79* ± 9.47	25.69* ± 8.49
BIA %FM SDS (n=47)	0.23 ± 0.98	0.35 ± 0.97	0.10 ± 1.00	0.37 ± 1.11	0.53 ± 1.08	0.25 ± 1.11
Ht^2/Impedance	30.24 ± 10.93	31.22 ± 13.31	29.15 ± 7.57			

Significantly different by Independent Samples Test *P< 0.05; **P< 0.001 Sig. (2-tailed). Standard deviation scores (SDS) for Wt, Ht, BMI, waist, and %FM were based on the UK90 reference data (Cole et al., 1998). BIA = Bioelectrical Impedance Analysis; SA = South Asian; Wt = weight; Ht = height; BMI = body mass index; WC = waist circumference; FM = fat mass; FFM = fat-free mass; SDS = standard deviation score

Figures 4.3 and 4.4 are respective scatterplots showing the distribution of the BMI SDS against age of the total study sample and sex-specific scatterplots of the external group. The scatterplots show that for both boys and girls, BMI SDS were evenly distributed around the mean, with very few subjects falling outside of the mean ± 3sd range.

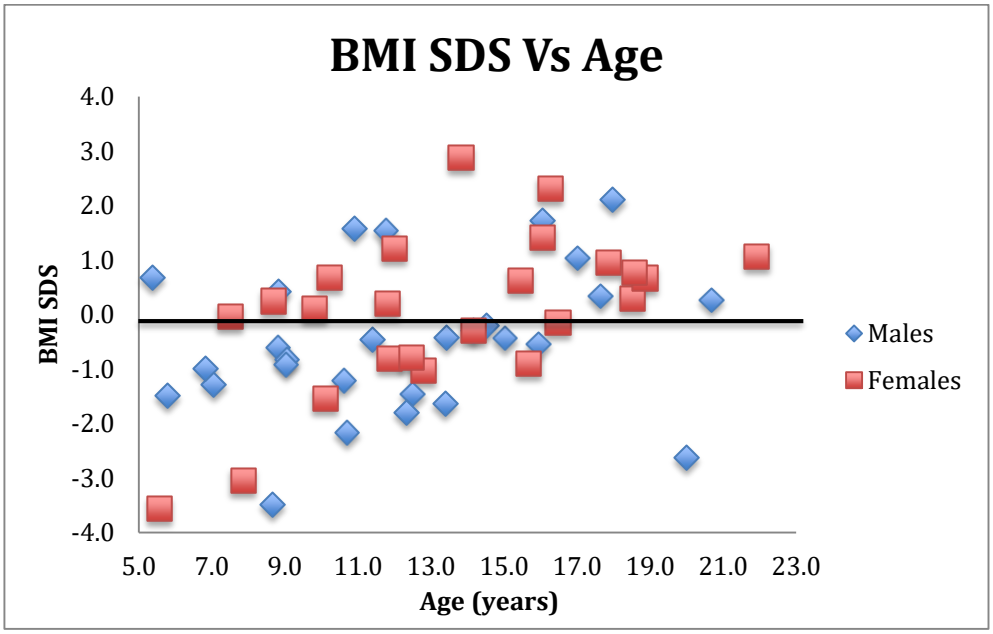
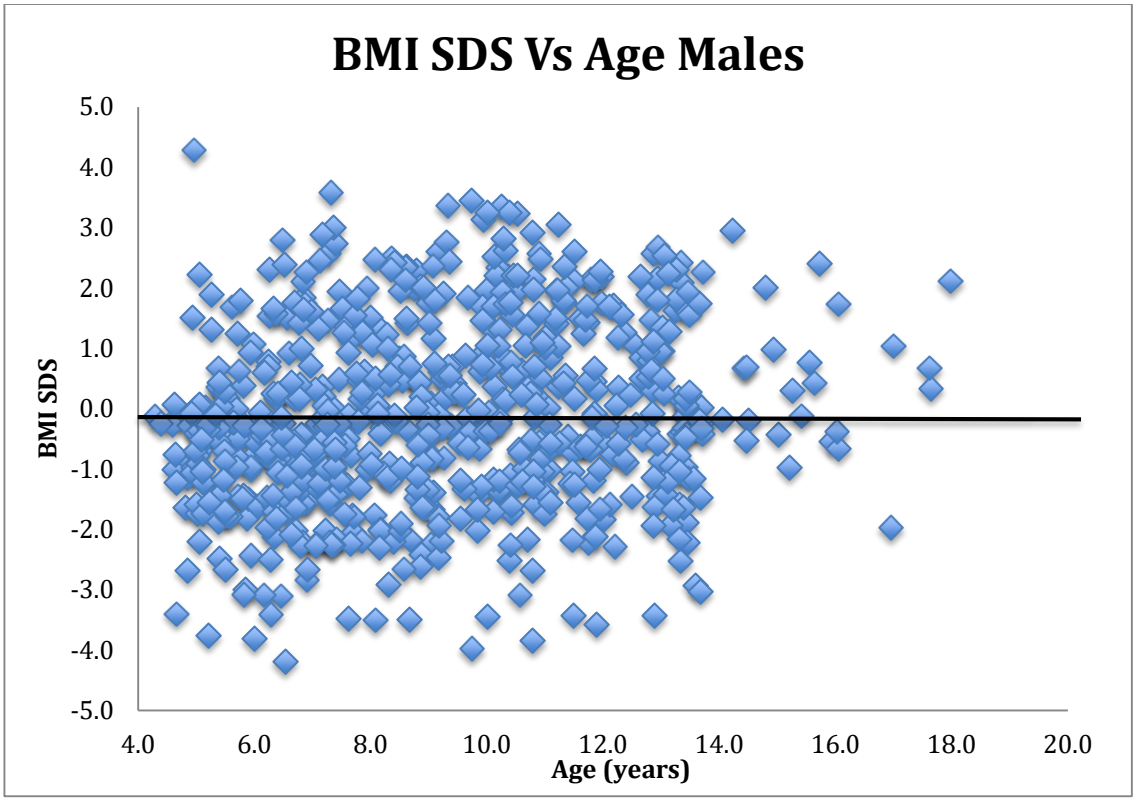


Figure 4.3 Scatterplot showing the distribution of the BMI SDS against age of the total study sample, based on the UK90 reference data. BMI = body mass index; SDS = standard deviation score



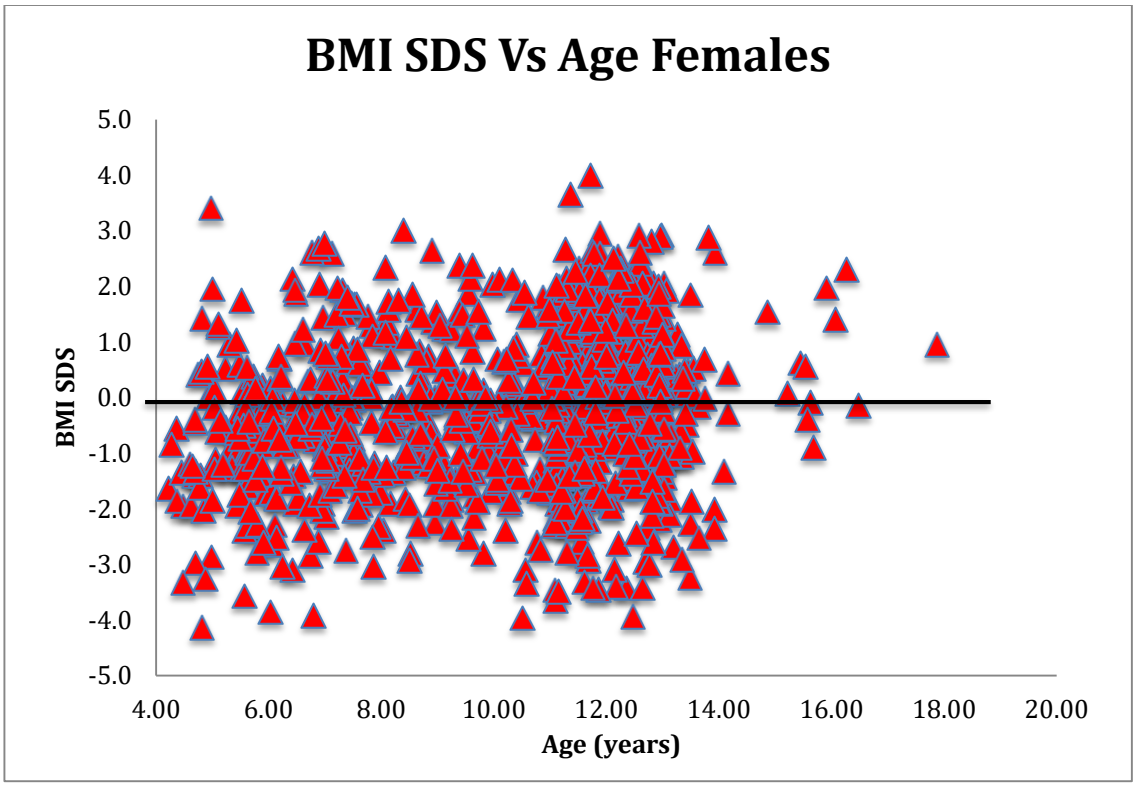


Figure 4.4 Scatterplot showing the distribution of the BMI SDS against age of the external South Asian group based on the UK90 reference data (upper graph = boys; lower graph = girls). BMI = body mass index; SDS = standard deviation score

4.5.2 Body Weight Comparisons

Both BIA and ADP Wt are determined by direct measurement using scale Wt. However, DXA Wt is determined by summing the weights of three component parts, i.e. BMC + soft-tissue FFM (LM) + FM. As there were no observed differences between BIA and ADP Wt, initial comparisons were made between DXA Wt and BIA Wt. Statistical comparisons between DXA and BIA Wt revealed that DXA Wt was strongly correlated with BIA Wt (R^2=998, r= 0.999; Figure 4.5). However, DXA consistently overestimated Wt compared to BIA (mean difference DXA – BIA = 2.47kg (sd 1.03); 95% limits of agreement (mean ±2sd) = 0.40 to 4.54kg (see Figure 4.6), which was significant at the P<0.001 level.

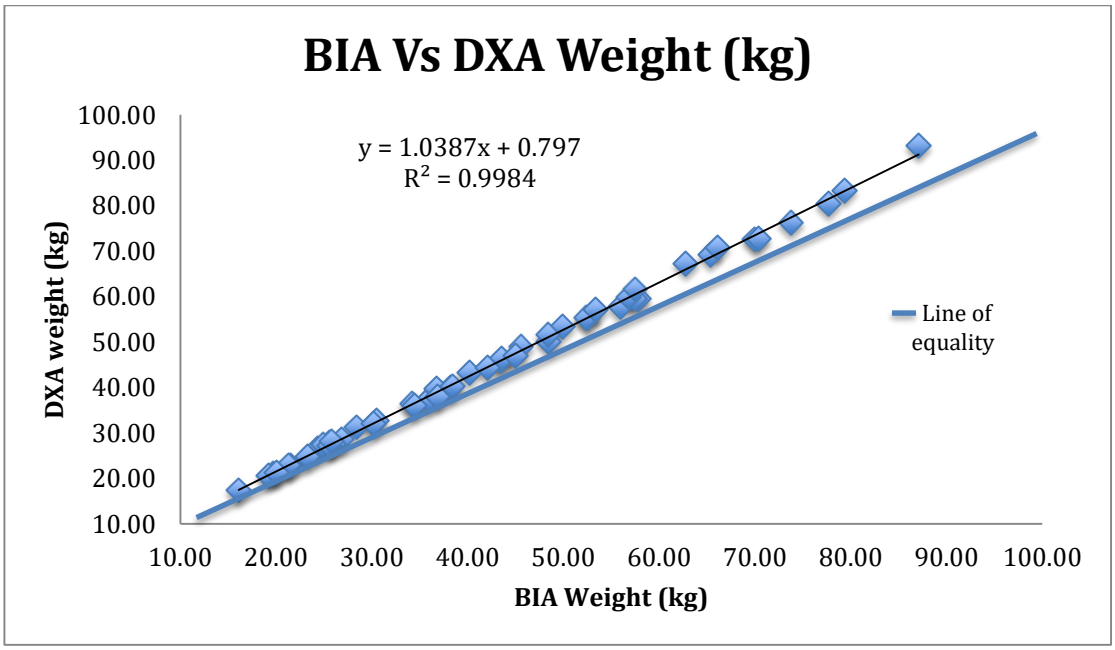


Figure 4.5 Scatterplot comparing BIA weight with DXA weight. BIA = bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry

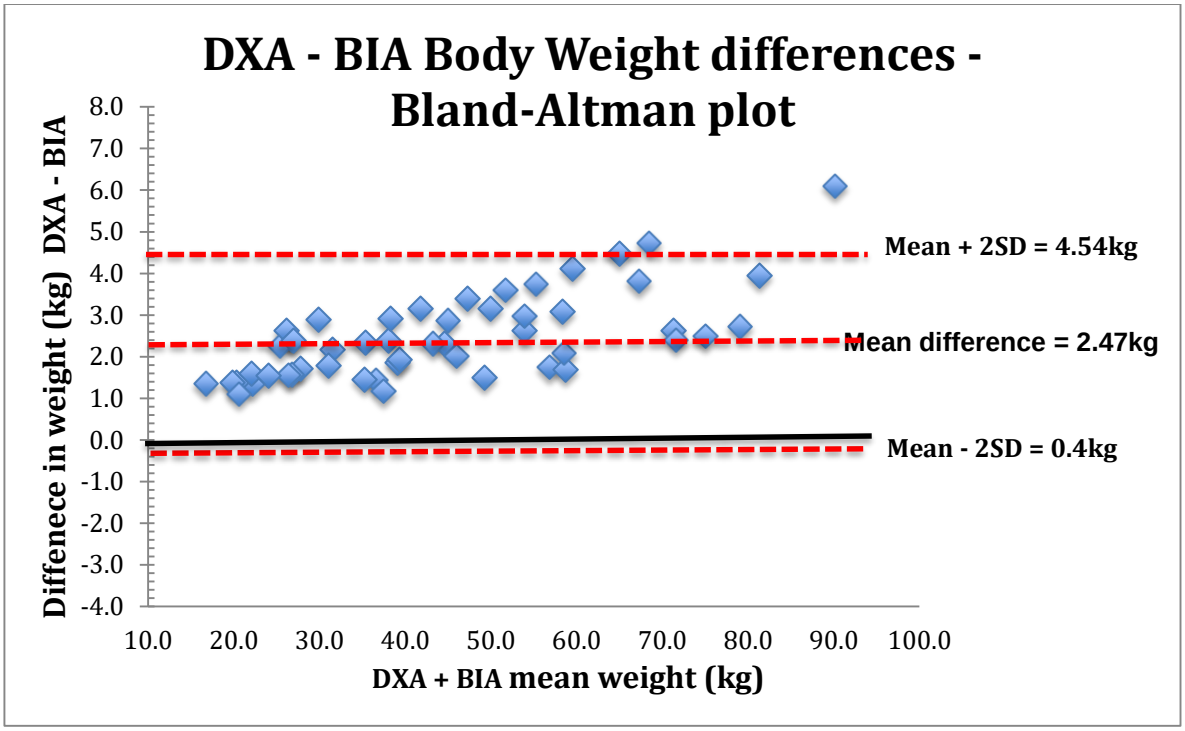


Figure 4.6 Bland-Altman comparison between DXA and BIA Weight. BIA =bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry

Due to the discrepancies in DXA Wt and scale Wt (BIA Wt), absolute values for DXA_{Siri} FM (kg) and FFM (kg), were determined in two different ways.

DXA_{Siri} FM:

i) DXA_{Siri} FM (kg)= DXA_{Siri} %FM x DXA Wt (i.e. BMC (kg) + DXA LM (kg) + DXA FM (kg)

ii) DXA_{Siri} FM2 (kg) = DXA_{Siri} %FM x BIA Wt (kg)

The first Siri FM (kg) value is represented as DXA_{Siri} FM, and the second Siri FM (kg) value, determined by using BIA scale Wt, is represented in all analyses as DXA_{Siri} FM2.

DXA_{Siri} FFM:

i) DXA_{Siri} FFM (kg) = (BMC (kg) + LM (kg) + FM (kg)) – DXA_{Siri} FM (kg)

ii) DXA_{Siri} FFM2 (kg) = DXA_{Siri} %FFM* x BIA Wt

* Based on the 2C model where Body Wt = FM +FFM, DXA_{Siri} %FFM was determined by subtracting DXA_{Siri} %FM from 100.

4.5.3 Body Composition Comparisons between BIA and DXA

Due to the significant gender differences for BIA FM and %FM (Table 4.1), body composition variables were analysed for the whole group and separately by gender. Table 4.2 provides body composition descriptive (mean) data for all absolute (kg) and relative (%) FM and FFM, for BIA and DXA, together with comparisons between the methods showing correlations, mean differences and limits of agreement.

Table 4.2: Body composition measures (mean ± sd) of SA BIA validation study group by BIA and DXA, for percentage fat-mass (%FM), absolute fat-mass (FM; kg), and absolute fat-free mass (FFM; kg), with Bland-Altman comparisons between methods showing mean difference and limits of agreement

	All (n =51)			**Boys (n =27)**			**Girls (n =24)**		
	Mean ±sd	**Mean difference ±sd (r) DXA - BIA**	**Limits of agreement (mean difference ± 2sd) DXA - BIA**	**Mean ±sd**	**Mean difference ±sd (r) DXA -BIA**	**Limits of agreement (mean difference ± 2sd) DXA - BIA**	**Mean ±sd**	**Mean difference ±sd (r) DXA -BIA**	**Limits of agreement (mean difference ± 2sd) DXA - BIA**
BIA FM (%)	21.82 ± 6.88	-	-	18.54 ± 4.96	-	-	24.94 ±5.91	-	-
DXA_{Siri} FM (%)	24.31± 9.06	2.49 ± 5.17, (0.82) *	-7.84 to 12.83	20.59 ± 7.86	2.23 ± 5.21, (0.77) *	-8.19 to 12.65	27.87 ±8.23	2.79 ± 5.21, (0.80) *	-7.64 to 13.22
DXA_{total} FM (%)	31.13 ±10.37	9.32 ± 6.33, (0.80) **	-3.35 to 21.98	26.52 ± 9.11	8.16 ± 6.36, (0.75) **	-	36.33 ±9.31	10.63 ± 6.19, (0.75) **	-
BIA FM (kg)	9.94 ±6.59	-	-	7.68 ± 4.16	-	-	12.47 ±7.89	-	-
DXA_{Siri} FM2¶ (kg)	10.87 ±6.93	0.93 ± 2.16, (0.95) *	-3.39 to 5.25	8.38 ± 4.70	0.70 ± 2.20, (0.88)	-3.70 to 5.10	13.66 ±8.01	1.18 ± 2.13, (0.96) *	-3.08 to 5.45
BIA FFM (kg)	33.29 ±13.29	-	-	33.86 ±15.96	-		32.66 ±9.77	-	
DXA_{Siri} FFM2¶ (kg)	32.26 ±13.50	-0.93 ± 2.16, (0.99) *	-5.25 to 3.39	33.16 ± 16.18	-0.70 ± 2.20, (0.99)	-5.10 to 3.70	31.48 ±9.92	-1.18 ± 2.13, (0.98) *	-5.45 to 3.08

SA = South Asian; BIA = Bioelectrical Impedance Analysis; DXA = Dual-energy x-ray absorptiometry; (r value = Paired Samples Correlations). Significant difference between BIA and DXA *P<0.05; **P< 0.001 Sig. (2-tailed Paired Samples Test). FM = fat mass; FFM = fat=free mass; ¶absolute DXA_{Siri} FM2 & FFM2 (kg), were weight corrected using BIA scale weight (see section 4.5.2).

4.5.3.1 %FM & %FFM comparisons between BIA & DXA

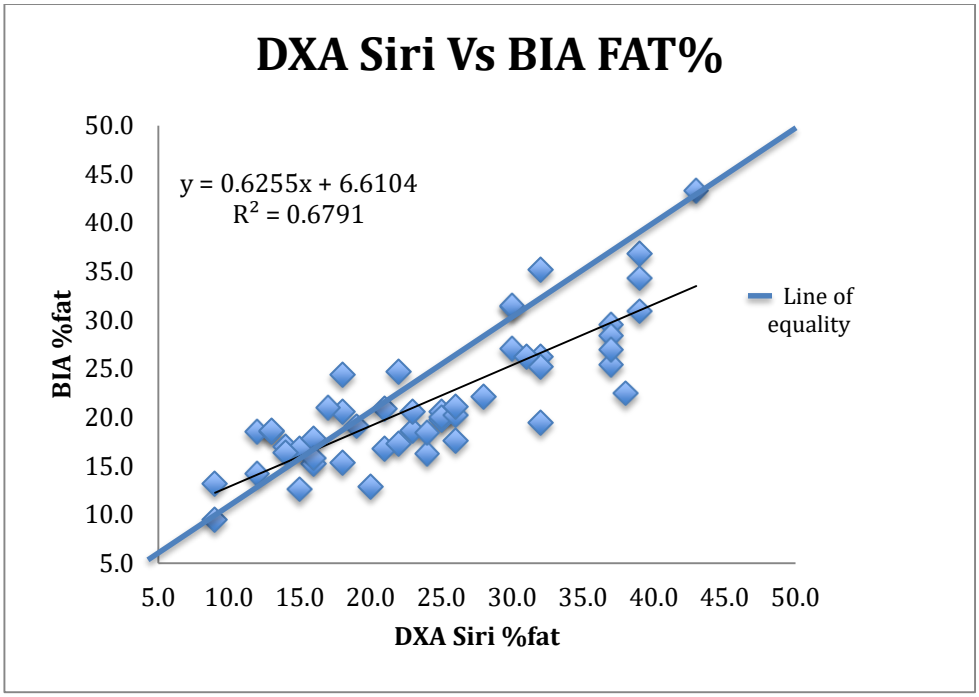
Initial comparisons were made between BIA %FM and both DXA_{Siri} and DXA_{total} %FM, to confirm that for this DXA model, as stipulated by the manufacturers, DXA_{Siri} was the appropriate output for body composition assessment. For completeness and confirmation that errors were not made in data entry, %FFM comparisons were also made, although were expected to be equivalent outputs of %FM, but opposite in direction.

Scatterplots showing the relationship between BIA and both DXA_{Siri} and DXA_{total} %FM are presented in Figures 4.7 to 4.9. The graphs show a strong positive correlation between BIA %FM and DXA (Siri and total) %FM, for the sample as a whole and for both sexes. The correlations were stronger between BIA %FM and DXA_{Siri} %FM (R^2 = 0.68, 0.59, 0.63), than between BIA %FM and DXA_{total} %FM (R^2= 0.65, 0.57, 0.56) for the whole sample and for boys and girls respectively. BIA %FM compared to DXA_{total} %FM almost consistently underestimated %FM. However, compared to DXA Siri %FM, BIA tended to overestimate %FM at the leaner end of the scale (<15% FM for boys, and <20%FM for girls), and underestimate fat at higher adiposity levels (Figure 4.8), with the level of underestimation increasing as %FM increased.

The paired-samples tests and Bland-Altman (Bland & Altman, 1986) comparisons revealed that for both DXA_{total} and DXA_{Siri} %FM, BIA significantly underestimated %FM (Table 4.2 and Figures 4.10 to 4.11). However, much larger differences were observed between BIA and DXA_{total} %FM than between BIA and DXA_{Siri} %FM. Compared to DXA_{total} %FM, BIA on average underestimated %FM by 9.32% (sd 6.33; P< 0.001) with limits of agreement (mean ± 2sd) ranging from -3.35 to 21.98% for the whole sample. However, comparisons between DXA_{Siri} and BIA %FM revealed that BIA on average underestimated %FM by 2.49% (sd 5.17; P = 0.001), with limits of agreement ranging from -7.84 to 12.83%.

Due to the large differences between BIA and DXA_{total} %FM, all other body composition comparisons were made between DXA_{Siri} variables only. Comparisons between BIA and DXA_{Siri} %FM separated by gender (Table 4.2 and Figure 4.11),

revealed that for boys and girls respectively, BIA on average significantly underestimated %FM by 2.23% (sd 5.21; P = 0.035; limits of agreement -8.19 to 12.65%), and 2.79% (sd 5.21; P = 0.015; limits of agreement -7.65 to 13.50%). As expected, equivalent outputs, in the opposite direction, were revealed for comparisons between BIA and DXA_{Siri} %FFM, i.e. in all cases compared to DXA_{Siri}, BIA significantly overestimated %FFM, by the equivalent amount it underestimated %FM (results not shown).



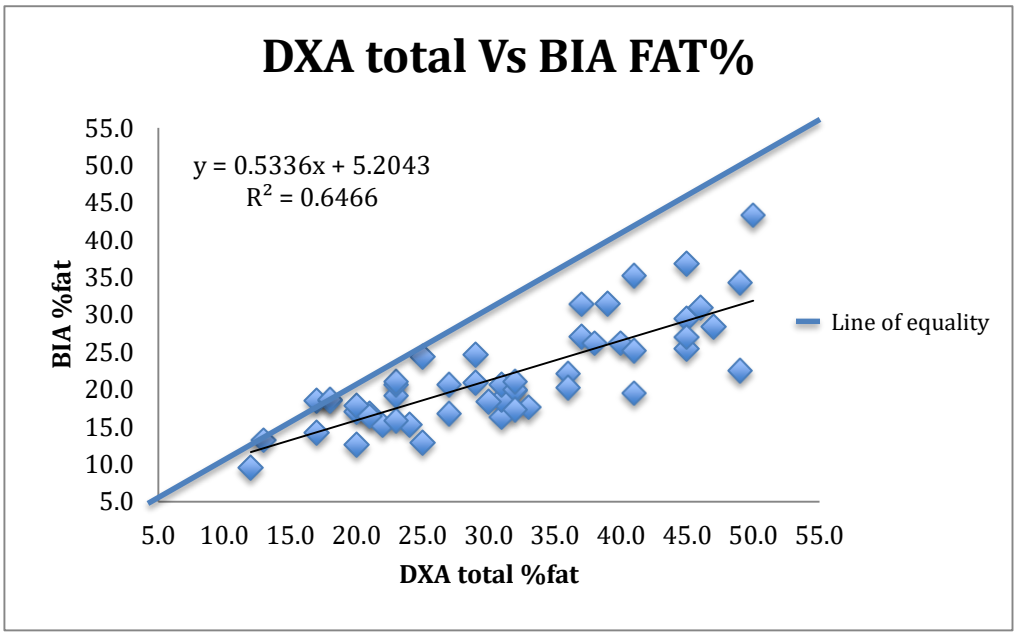
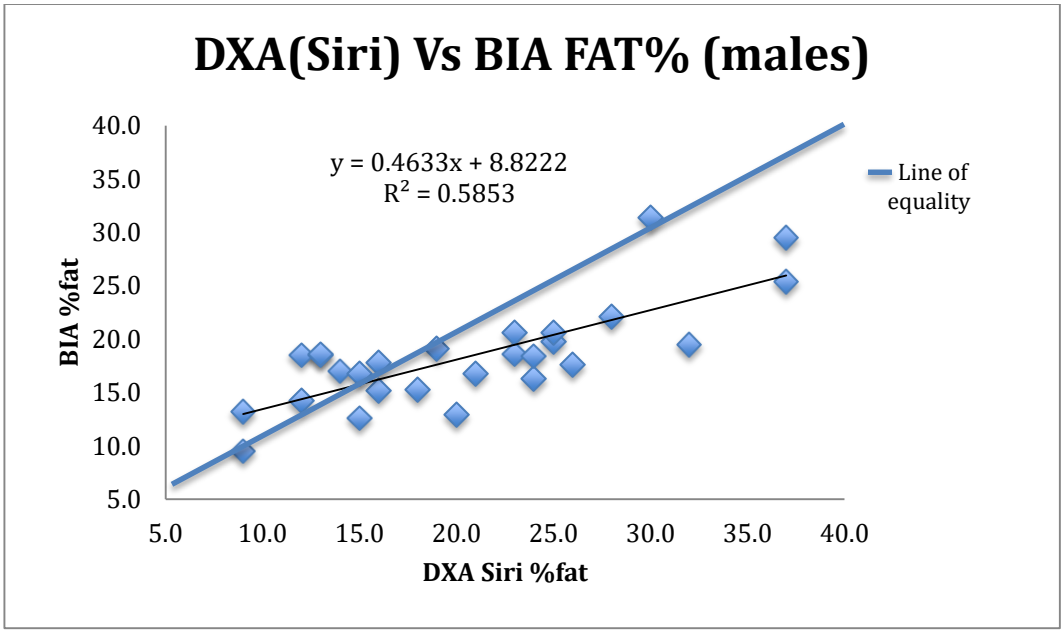


Figure 4.7 Scatterplots comparing BIA %FM with DXASiri (upper graph; BIA %FM = 0.63 x DXASiri %FM + 6.6) and DXAtotal %FM (lower graph; BIA %FM = 0.53 x DXAtotal %FM + 5.2) for whole sample. FM = fat mass; BIA =bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry. DXASiri & DXAtotal outputs (see section 4.3.3).



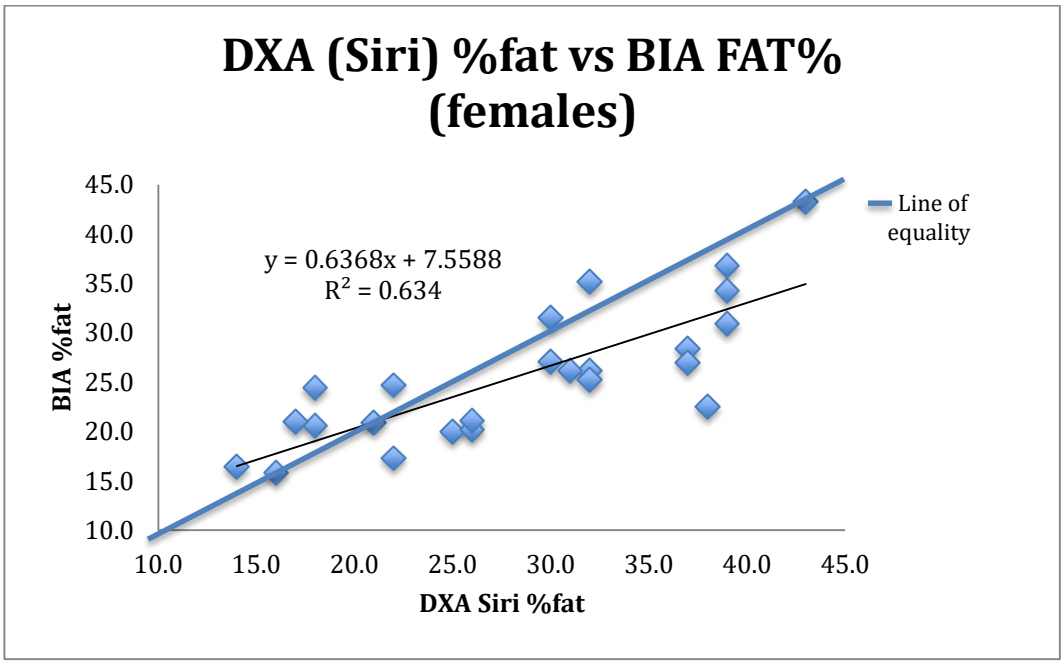
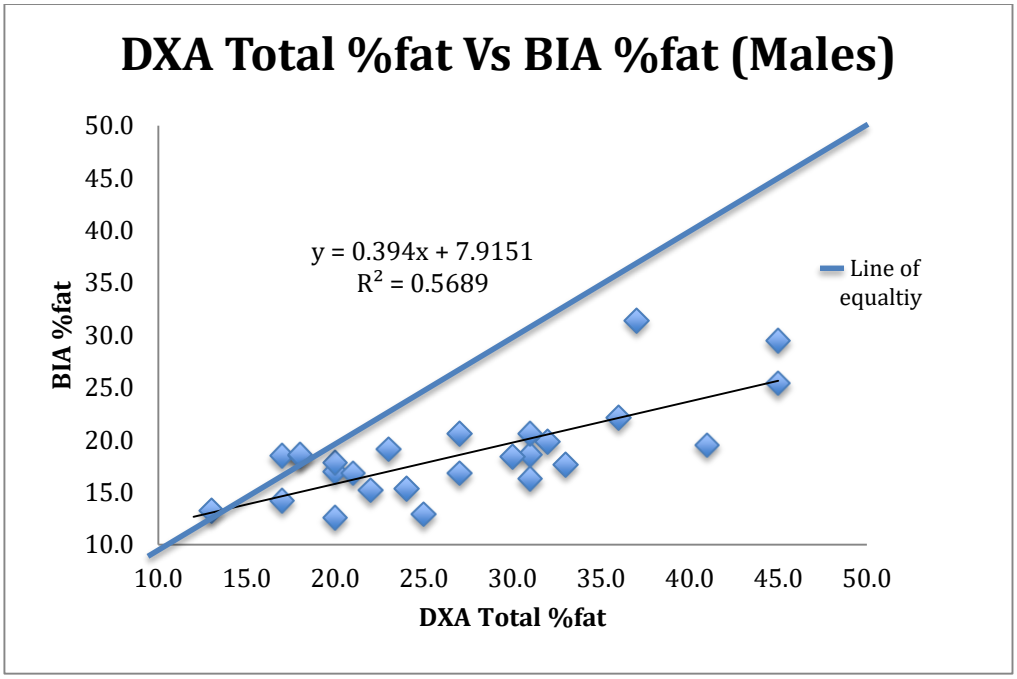


Figure 4.8 Scatterplots comparing BIA %FM with DXASiri %FM (upper graph boys only and lower graph girls only). FM = fat mass; BIA =bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry. DXASiri & DXAtotal outputs (see section 4.3.3).



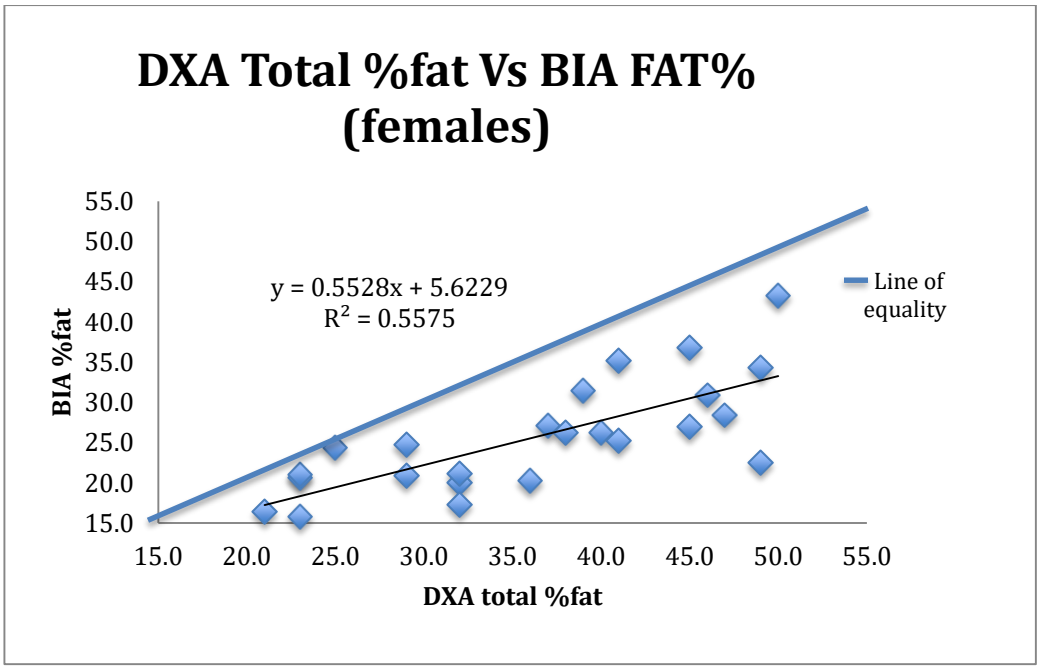
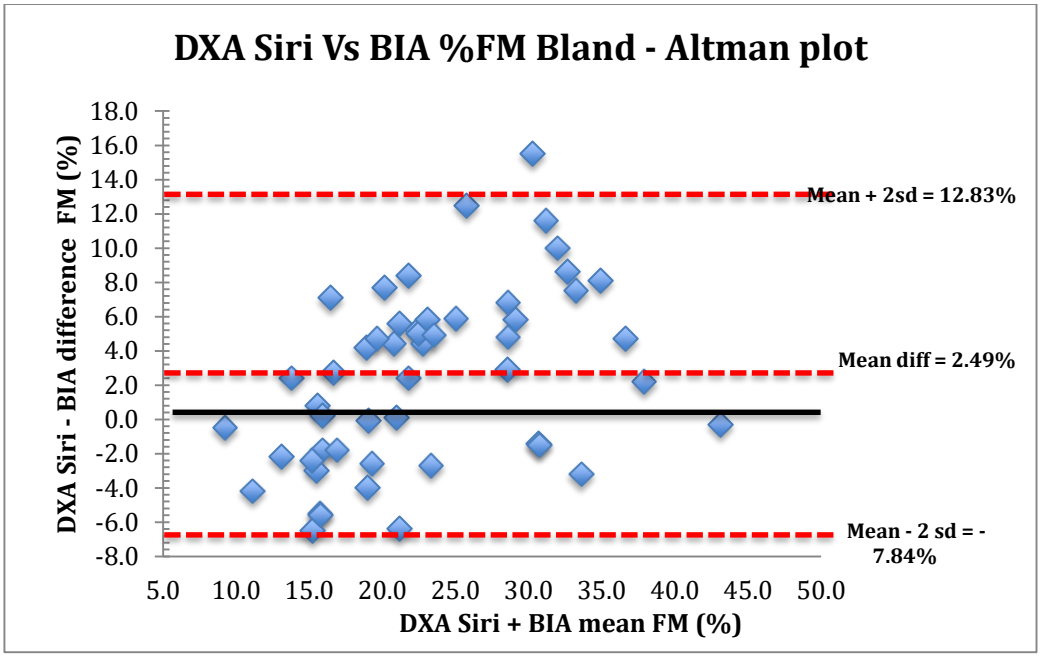


Figure 4.9 Scatterplots comparing BIA %FM with DXAtotal %FM (upper graph boys only and lower graph girls only). FM = fat mass; BIA =bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry. DXASiri & DXAtotal outputs (see section 4.3.3).



FM (%)

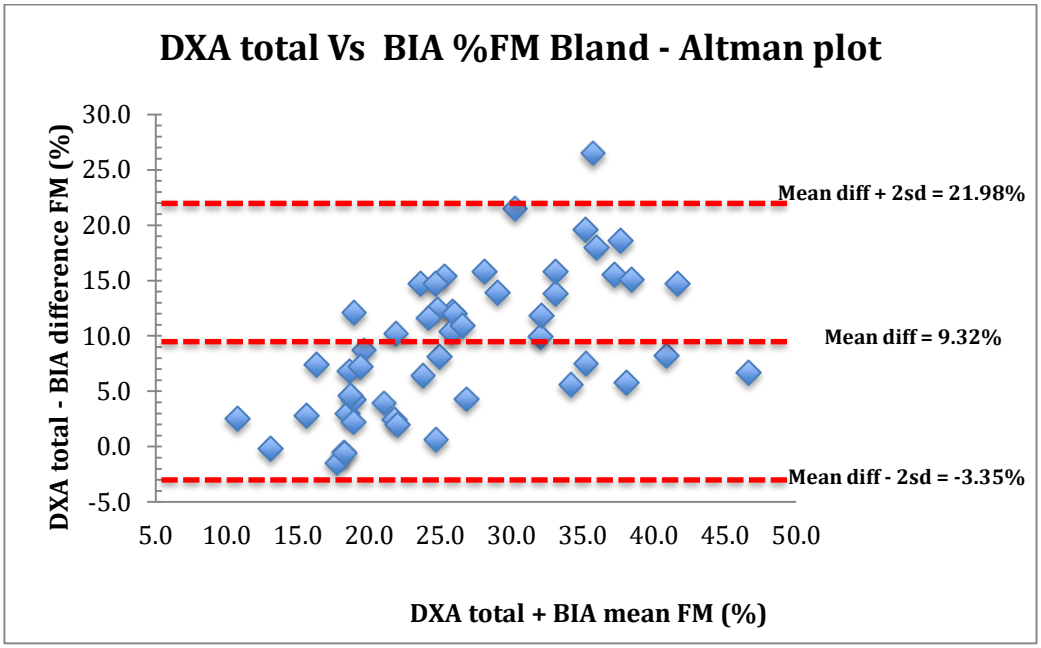
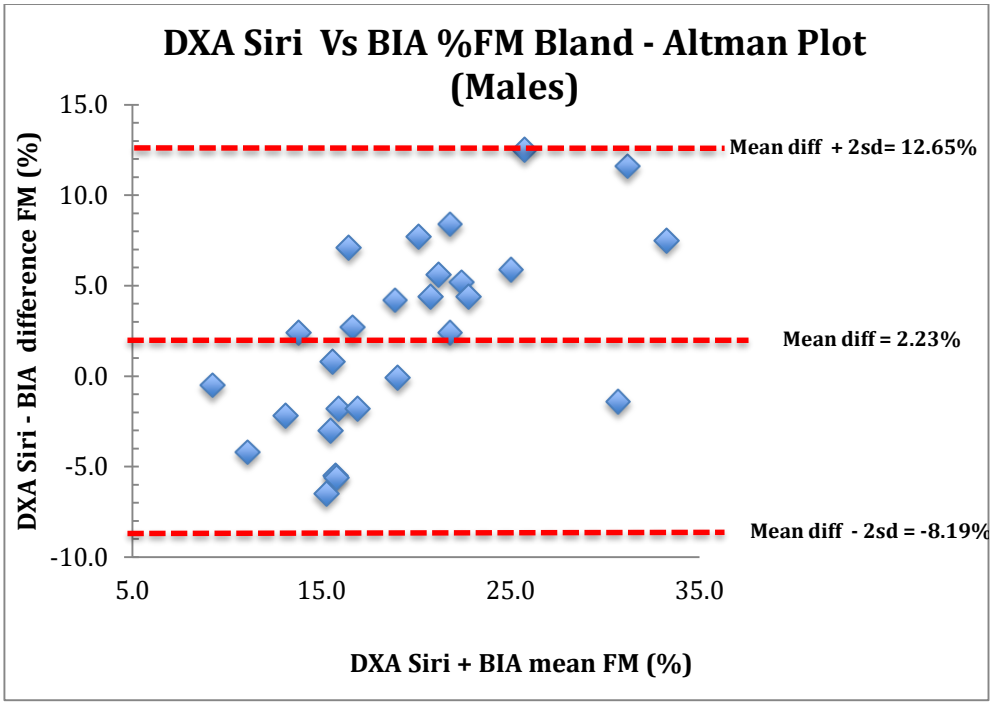


Figure 4.10 Bland–Altman plot showing mean difference & limits of agreement (mean ±2sd) between BIA %FM with DXASiri (upper graph) and between DXAtotal %FM (lower graph) for whole sample. FM = fat mass; BIA =bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry. DXASiri & DXAtotal outputs (see section 4.3.3).



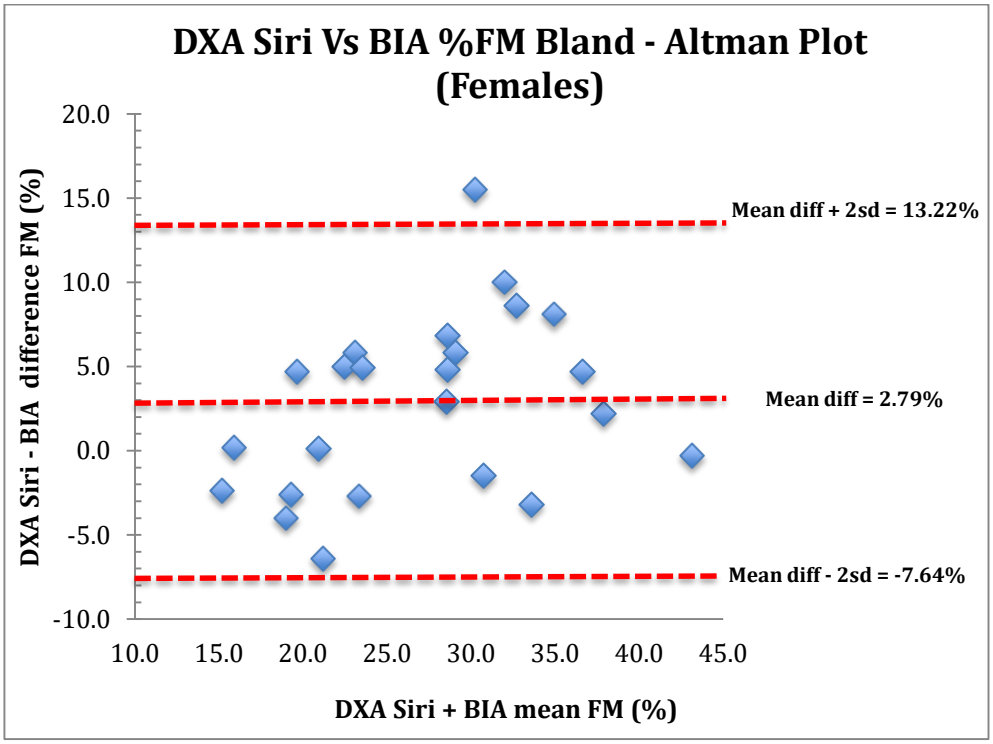
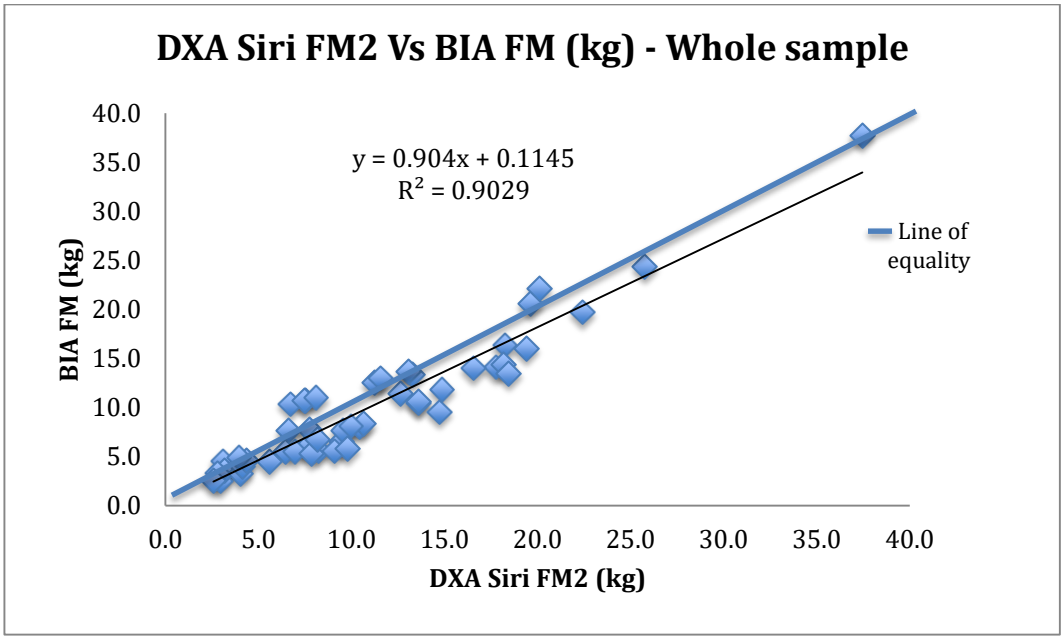


Figure 4.11 Bland–Altman plot showing mean difference & limits of agreement (mean difference ±2sd) between DXASiri and BIA %FM (upper graph boys only, lower graph girls only). FM = fat mass; BIA =bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry. DXASiri & DXAtotal outputs (see section 4.3.3).

4.5.3.2 FM & FFM comparisons between BIA & DXA

In all cases BIA strongly correlated with DXA (i.e. FM2 and FFM2) for both FM (R^2 = 0.90; 0.78; 0.93) and FFM (R^2 = 0.97; 0.98; 0.95), for the whole sample and boys and girls respectively (Figures 4.12 to 4.14). The mean difference and limits of agreement for FM and FFM between BIA and DXA as expected gave equivalent outputs, but in the reverse direction (Figures 4.15 to 4.17 and Table 4.2). Compared to DXA, BIA underestimated FM and overestimated FFM. This difference was significant in the whole sample (FM mean difference = 0.93kg; sd = 2.16; limits of agreement = -3.39 to 5.25kg; P = 0.003), and in the female sample (FM mean difference = 1.18kg; sd 2.13; limits of agreement = -3.08 to 5.45kg; P = 0.012). However, the difference between BIA and DXA_{Siri} for FM2 and FFM2, in the male sample, was not significant (FM mean difference = 0.70kg; sd = 2.20; limits of agreement = -3.70 to 5.10; P = 0.108).



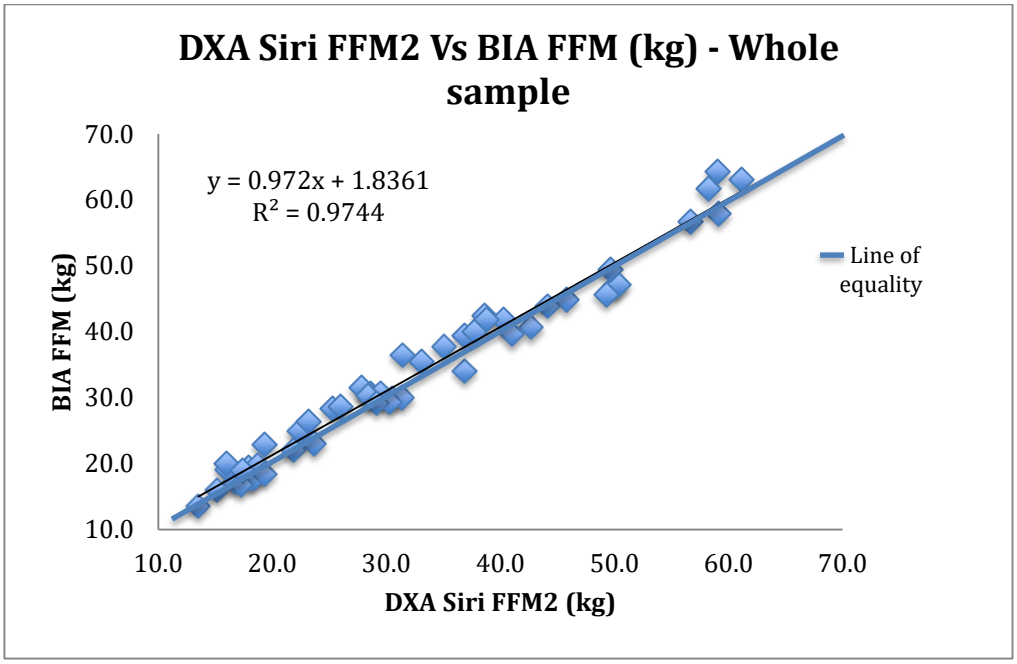
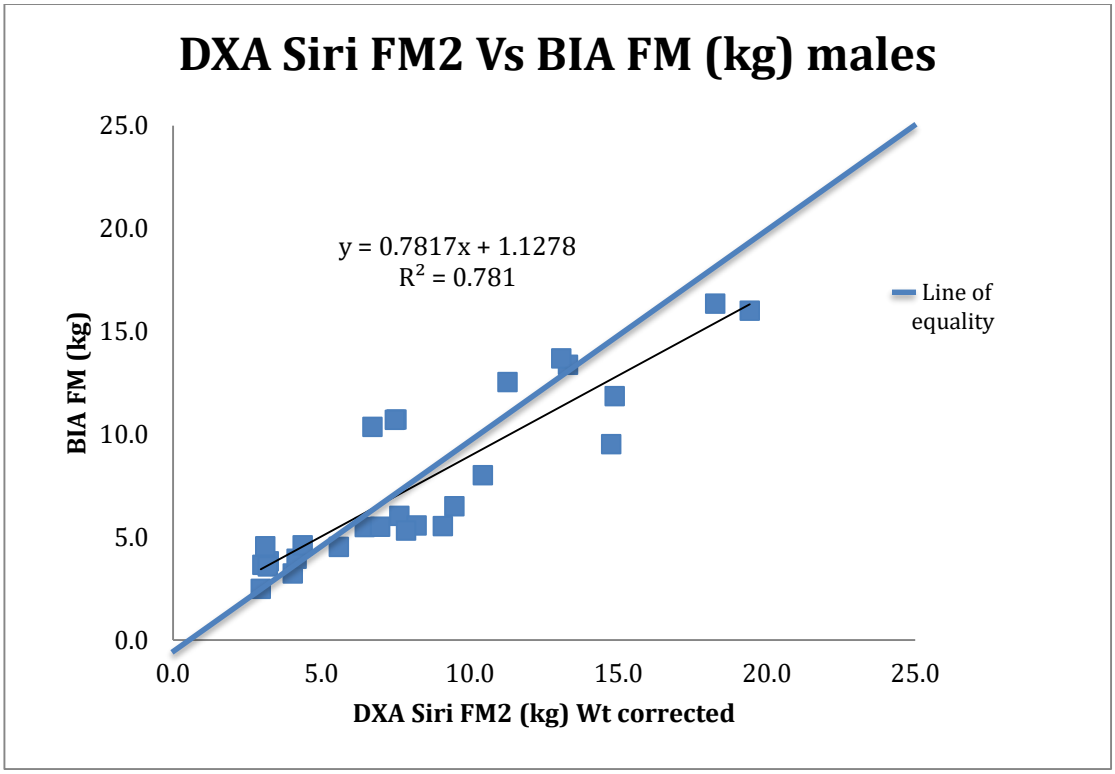


Figure 4.12 Scatterplots comparing BIA FM with DXASiri FM2 (kg; upper graph) and BIA FFM with DXASiri FFM2 (kg; lower graph) for whole sample. DXASiri FM2 & FFM2 were weight (Wt) corrected using scale Wt (see section 4.5.2). FM = fat mass; FFM = fat-free mass; BIA =bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry.



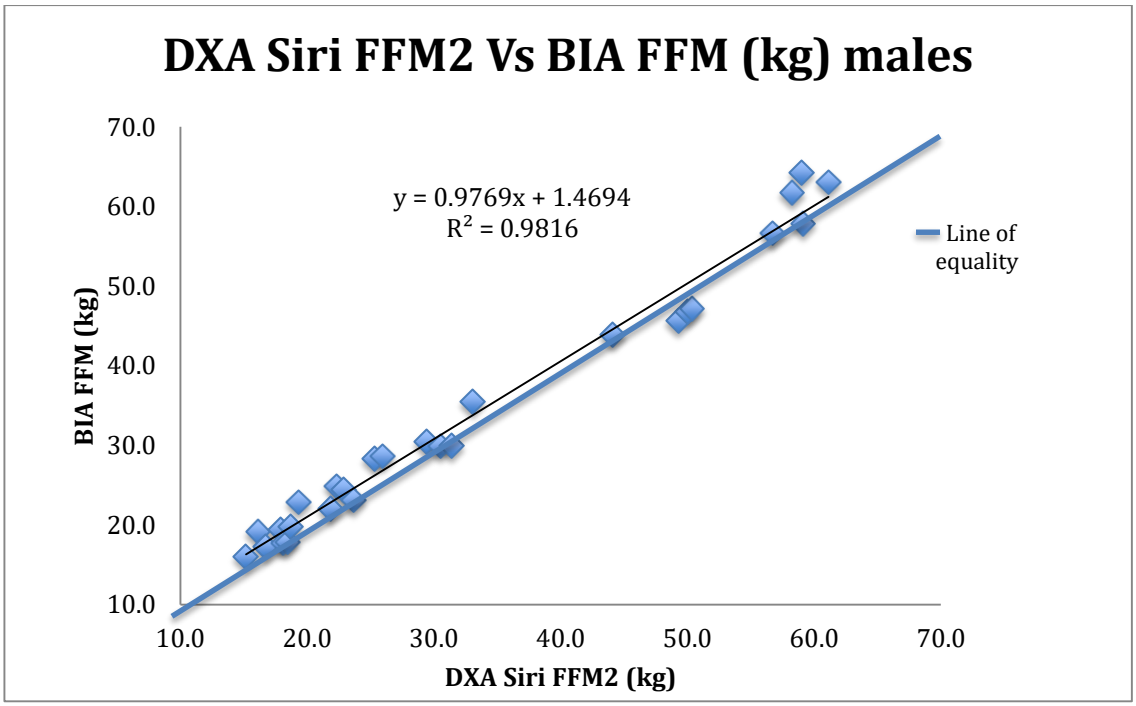
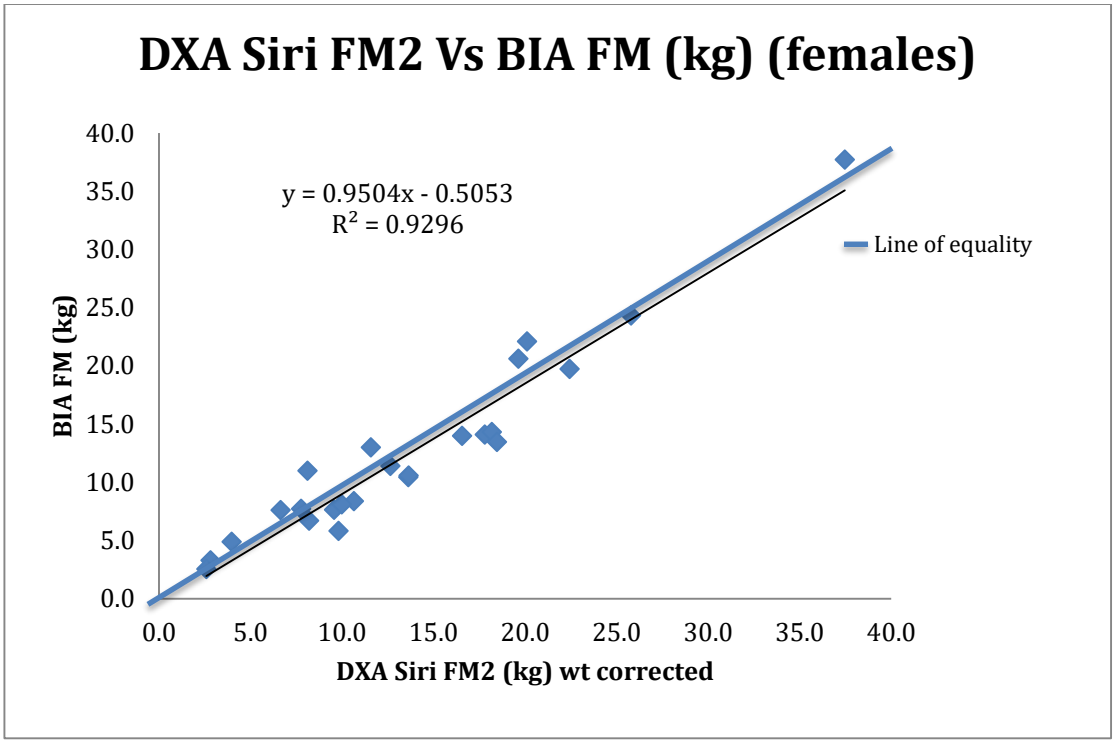


Figure 4.13 Scatterplots comparing BIA FM with DXASiri FM2 (kg; upper graph) and BIA FFM with DXASiri FFM2 (kg; lower graph) for boys only. DXASiri FM2 & FFM2 were weight (Wt) corrected using scale Wt (see section 4.5.2). FM = fat mass; FFM = fat-free mass; BIA = bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry



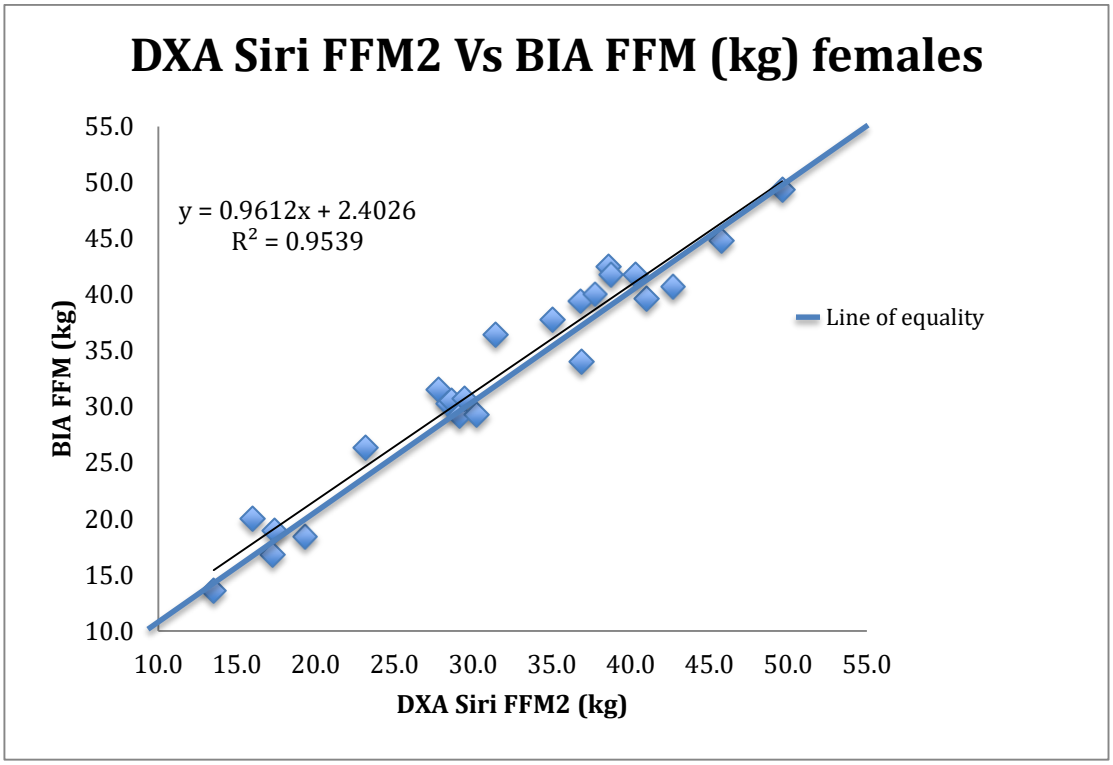
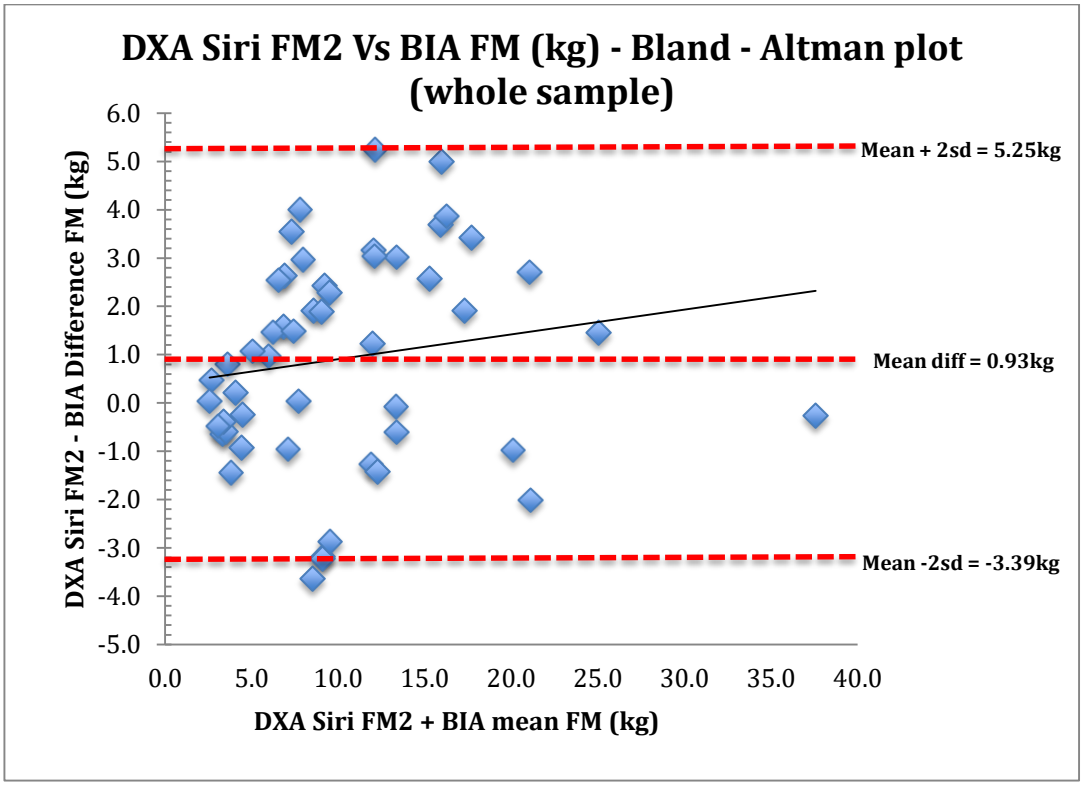


Figure 4.14 Scatterplots comparing BIA FM with DXASiri FM2 (kg; upper graph) and -BIA FFM with DXASiri FFM2 (kg; lower graph) for girls only. DXASiri FM2 & FFM2 were weight (Wt) corrected using scale Wt (see section 4.5.2). FM = fat mass; FFM = fat-free mass; BIA = bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry



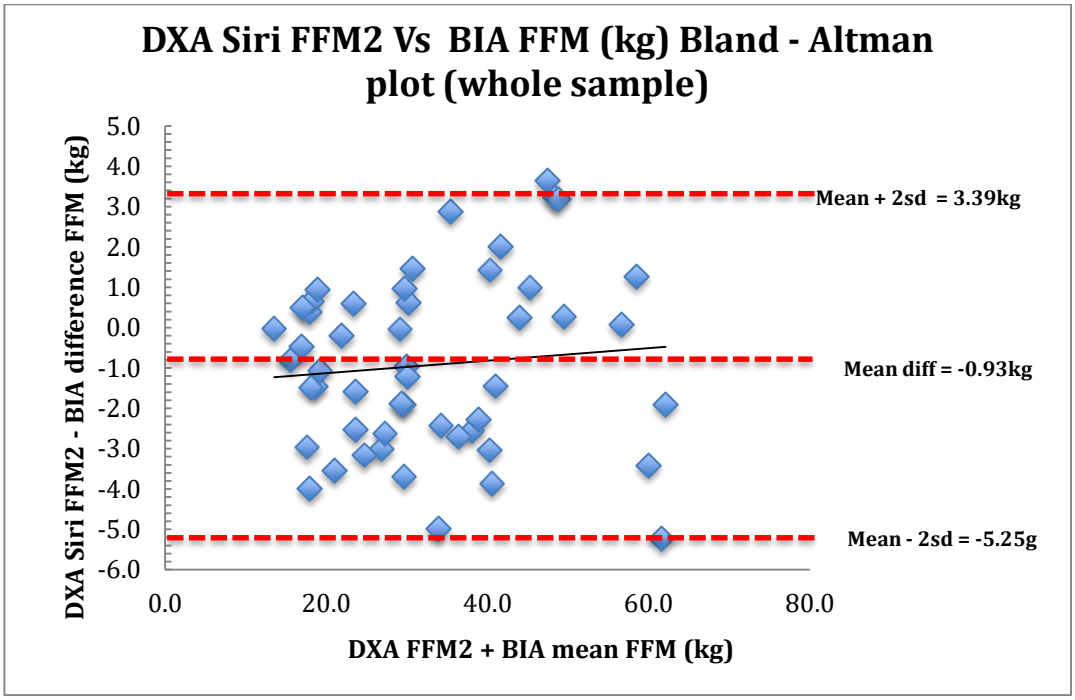
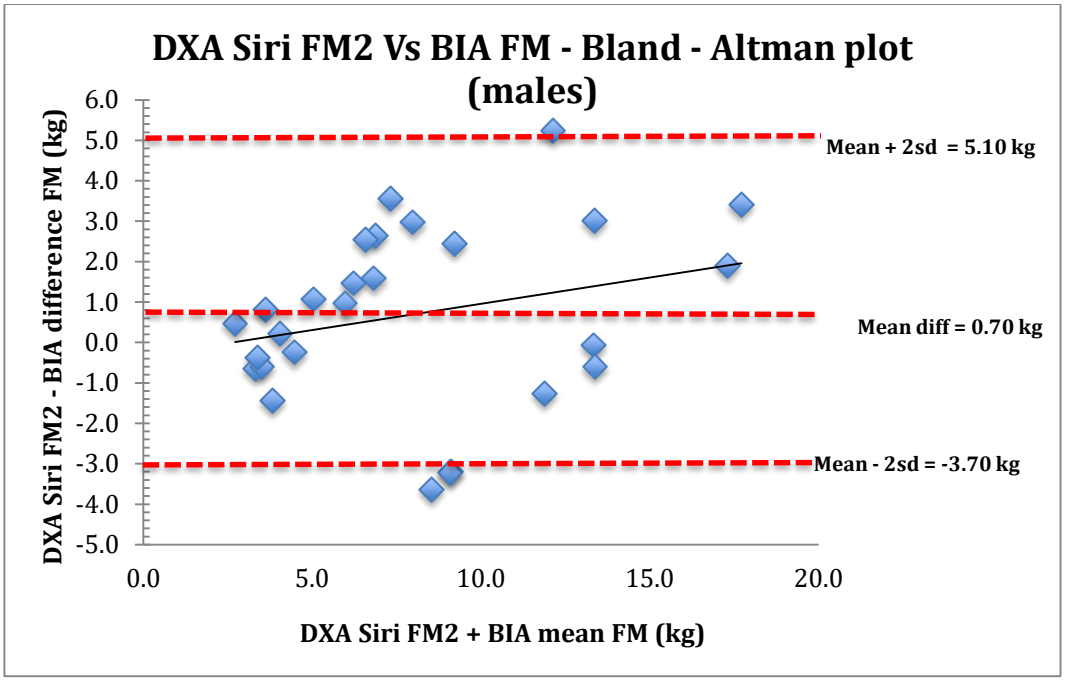


Figure 4.15 Bland–Altman plot showing mean difference & limits of agreement (mean difference + 2sd) between BIA FM with DXASiri FM2 (upper graph) and between DXASiri FFM2 (lower graph) for whole sample. DXASiri FM2 & FFM2 were weight (Wt) corrected using scale Wt (see section 4.5.2). FM = fat mass; FFM = fat-free mass; BIA = bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry



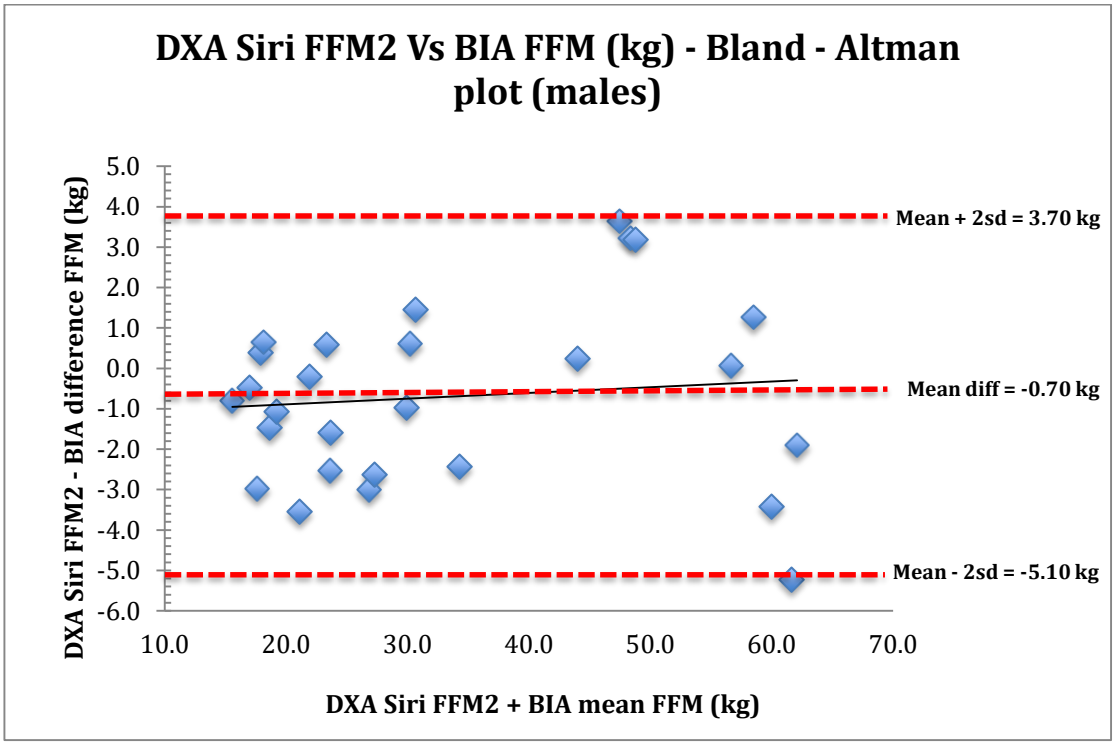
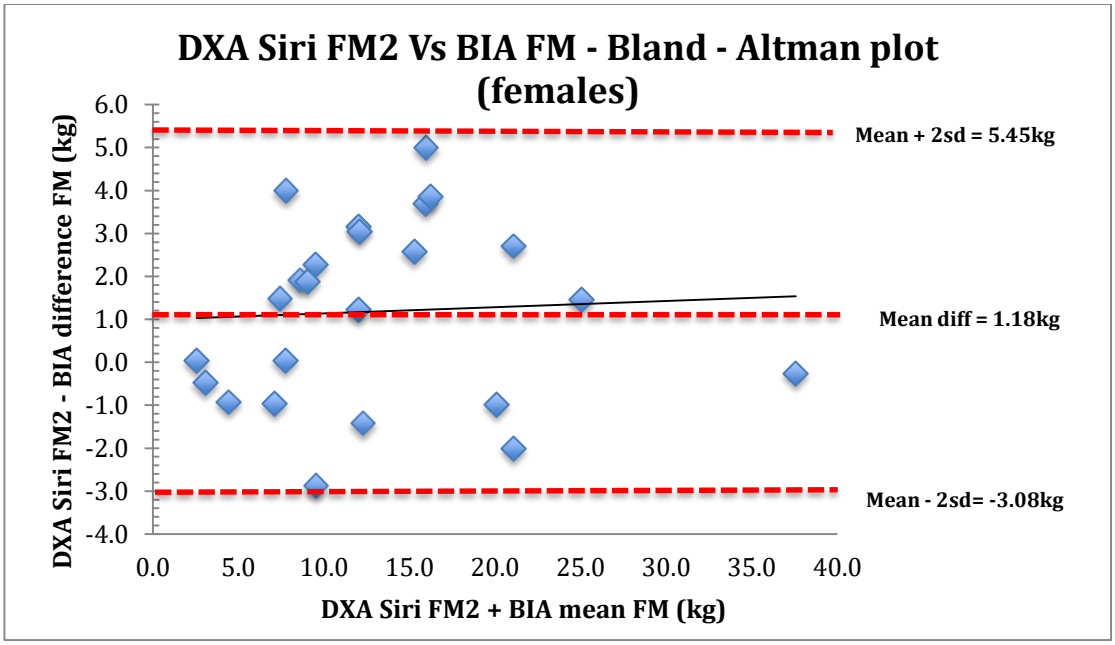


Figure 4.16 Bland – Altman plot showing mean difference & limits of agreement (mean difference + 2sd) between BIA FM with DXASiri FM2 (upper graph) and between DXASiri FFM2 (lower graph) for boys only. DXASiri FM2 & FFM2 were weight (Wt) corrected using scale Wt (see section 4.5.2). FM = fat mass; FFM = fat-free mass; BIA = bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry



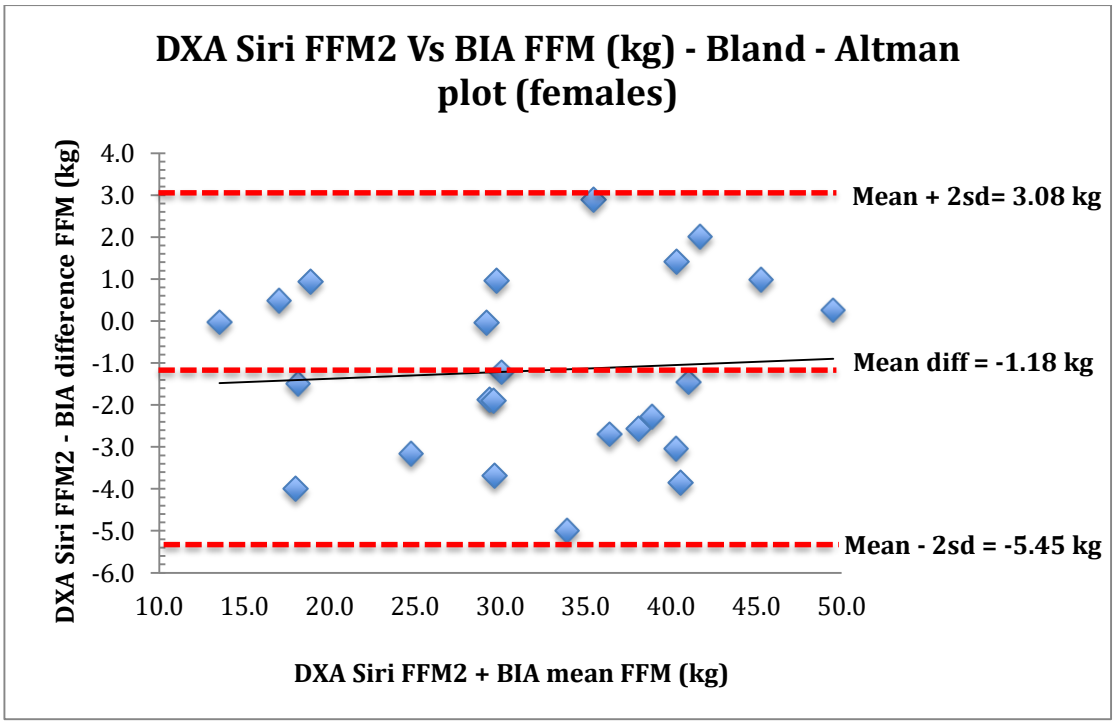
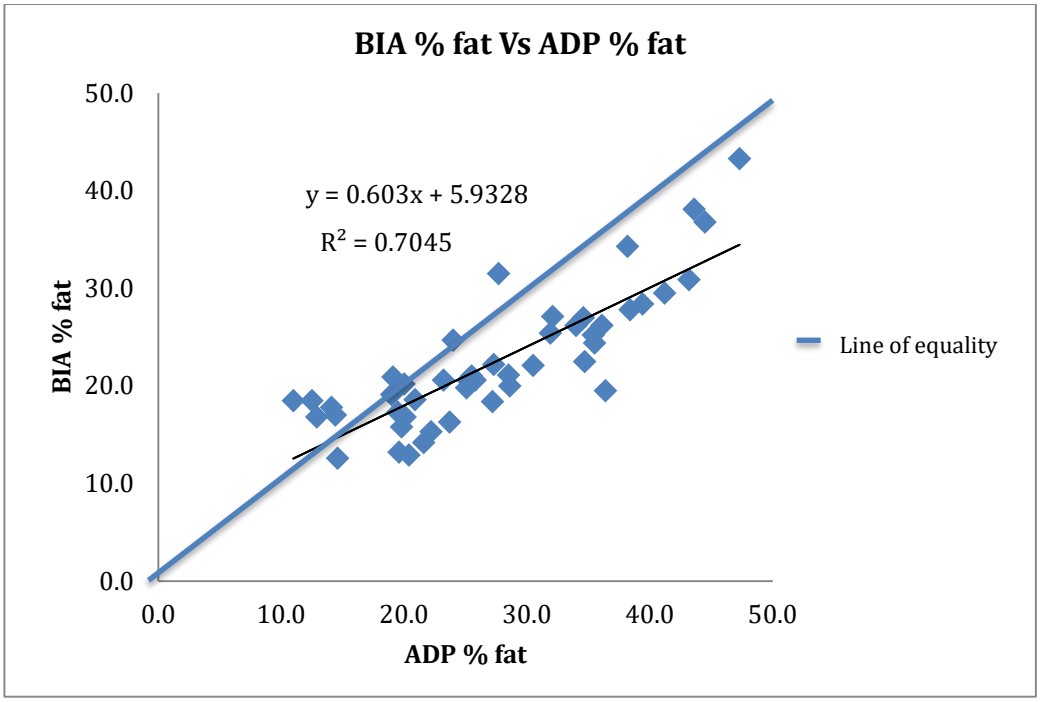


Figure 4.17 Bland–Altman plot showing mean difference & limits of agreement (mean difference + 2sd) between BIA FM with DXASiri FM2 (upper graph) and between DXASiri FFM2 (lower graph) for girls only. DXASiri FM2 & FFM2 were weight (Wt) corrected using scale Wt (see section 4.5.2). FM = fat mass; FFM = fat-free mass; BIA = bioelectrical impedance analysis; DXA = dual-energy x-ray absorptiometry

4.5.3.3 ADP comparisons with BIA & DXA

Similar to the DXA outcomes reported above, BIA consistently underestimated %FM compared to ADP as can be seen by the line of equality in the upper graph in Figure 4.18 showing a strong positive correlation between BIA and ADP ($r = 0.84$, $r^2 = 0.71$). The Bland-Altman plot also revealed a mean difference between ADP and BIA (BIA - ADP) of -4.9% with the 95% limits of agreement ranging from -14.8% to 5%. As seen in Figure 4.19, a strong positive relationship was observed between DXA and ADP for % fat ($r=0.87$, $r^2 = 0.75$); however, there did appear to be one outlier, and removal of this data point improved the relationship ($r=0.90$, $r^2 = 0.81$), and the mean difference between the two measures fell slightly to 1.2%. However, a significant difference was observed in %FM between DXA and ADP with the outlier included ($p = 0.03$) with a mean difference (ADP – DXA) between the two methods of 1.5%, with ADP having an overall tendency to underestimate %FM. Although removal of the outlier reduced the difference slightly, this remained significant ($p=0.05$). Whilst the mean difference between these two methods was small (Bland-Altman graph Figure 4.19), the 95% limits of agreement (mean ± 2sd) were very large, ranging from -18.8% to 21.2 %FM, with 4 data points falling outside this range.



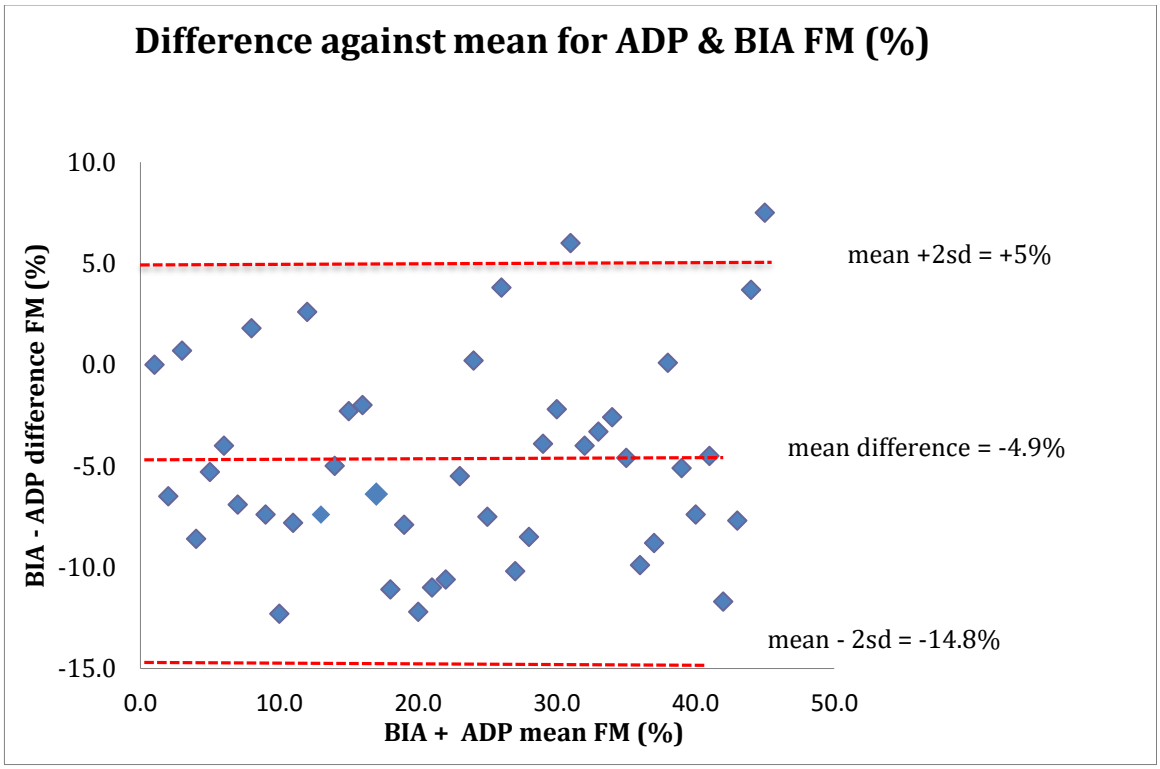
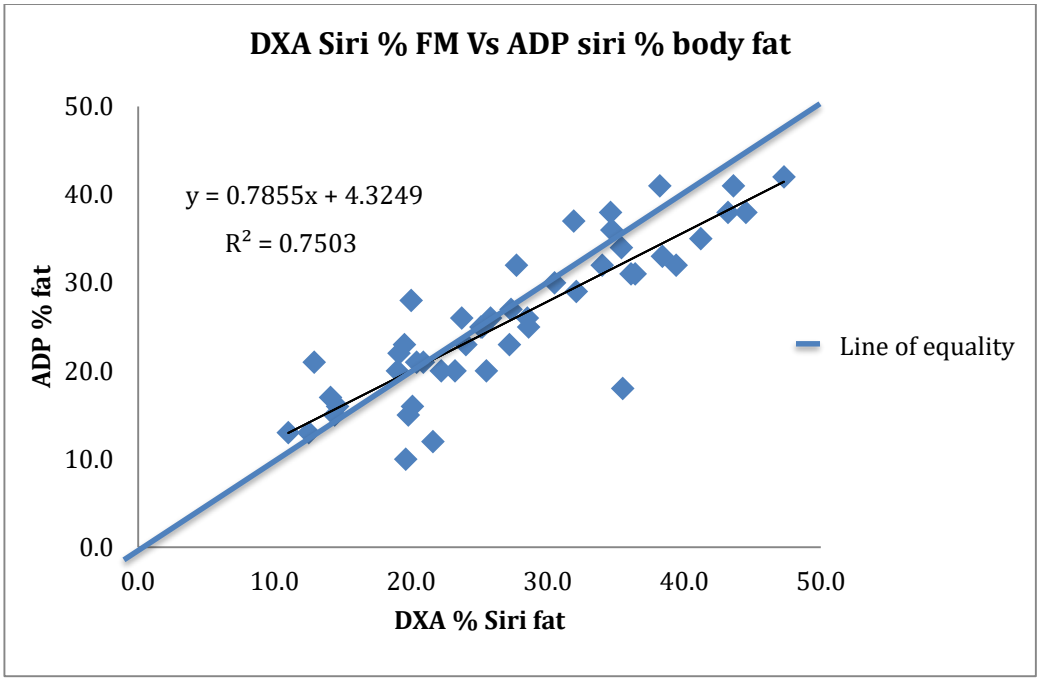


Figure 4.18 Upper graph correlation of BIA whole body % fat vs ADP whole body % fat (%FMBIA = 0.6 x %FMBodPod + 5.9; r = 0.84, P<0.001) with line of equality. Lower graph Bland-Altman plot showing limits of agreement between %FM for BIA and ADP. FM = fat mass; BIA = bioelectrical impedance analysis; ADP = air displacement plethysmography



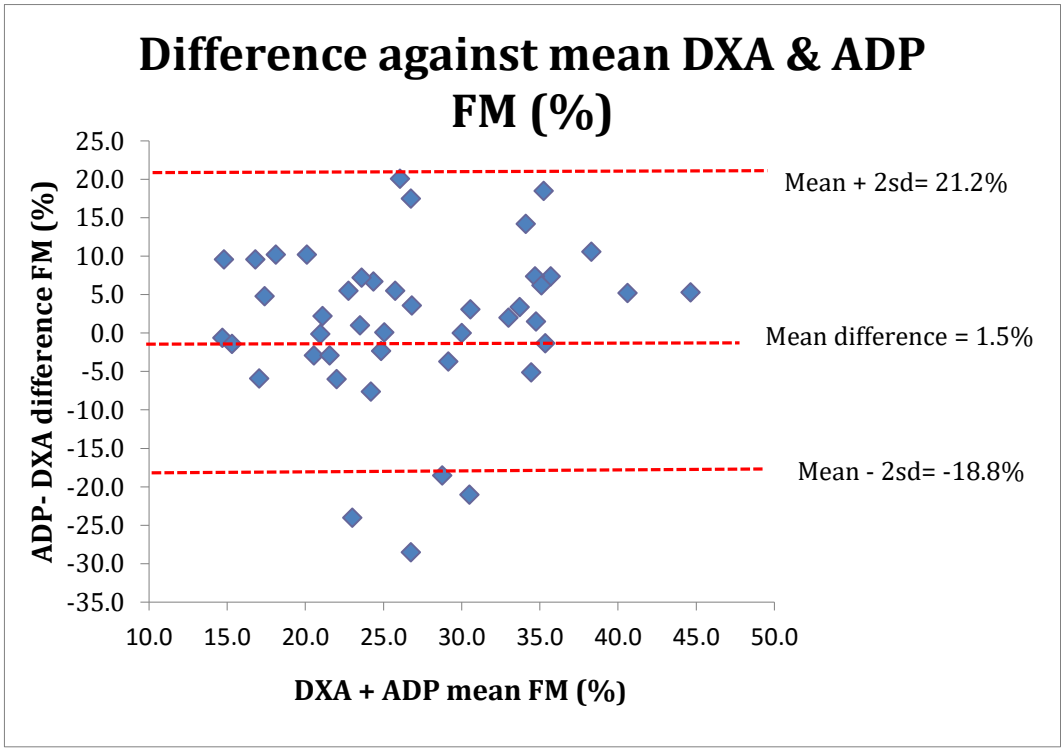


Figure 4.19 Upper graph correlation of DXA whole body % fat vs ADP whole body % fat (%FMBodPod = 0.8 x %FMDXA+ 4.3; r = 0.87, P<0.001) with line of equality. Lower graph Bland-Altman plot showing limits of agreement between %FM for DXA and ADP. FM = fat mass; DXA = dual-energy x-ray absorptiometry; ADP = air displacement plethysmograph; DXAsiri (see section 4.3.3).

Due to the significant differences in %FM between BIA and ADP, and ADP and DXA, with very large limits of agreement, it seemed likely that ADP was producing some spurious results, thus further analyses were conducted solely between BIA and DXA,

and given that the in-built BIA equation for this model was validated against method, it seemed prudent to use the same method.

4.5.3.4 %FM, absolute (kg) FM and FFM comparisons with different %FM and age cut-offs

Due to observed variations in the scatterplots, with BIA overestimating %FM for the leaner sample, and underestimating %FM for subjects with higher adiposity (see Figures 4.7 to 4.8), more detailed analyses were carried out on BIA %FM, absolute FM (kg) and absolute FFM (kg), separating the sample by gender and using a %FM cut-off based on DXA_{Siri} %FM, and secondly an age cut-off, which roughly divided the sample in half.

For boys, the two %FM cut-offs were DXA Siri %FM ≤ 20% and >20%; and for girls the two cut-offs were DXA Siri %FM ≤ 30% and >30%. The scatterplots for both %FM cut-offs are presented in Figures 4.20 to 4.23. Results for paired-samples tests comparing BIA %FM with DXA_{Siri} %FM, are presented in Table 4.3. No significant differences were observed between BIA %FM and DXA_{Siri} %FM for the leaner samples for either gender with BIA only slightly overestimating %FM compared to DXA Siri, (mean difference = -1.3%; p = 0.217 for boys; and mean difference = -0.31%; p = 0.786 for girls). However, the correlations were weak (r = 0.28; R^2 = 0.079) for boys, and strong (r = 0.67; R^2 = 0.45).

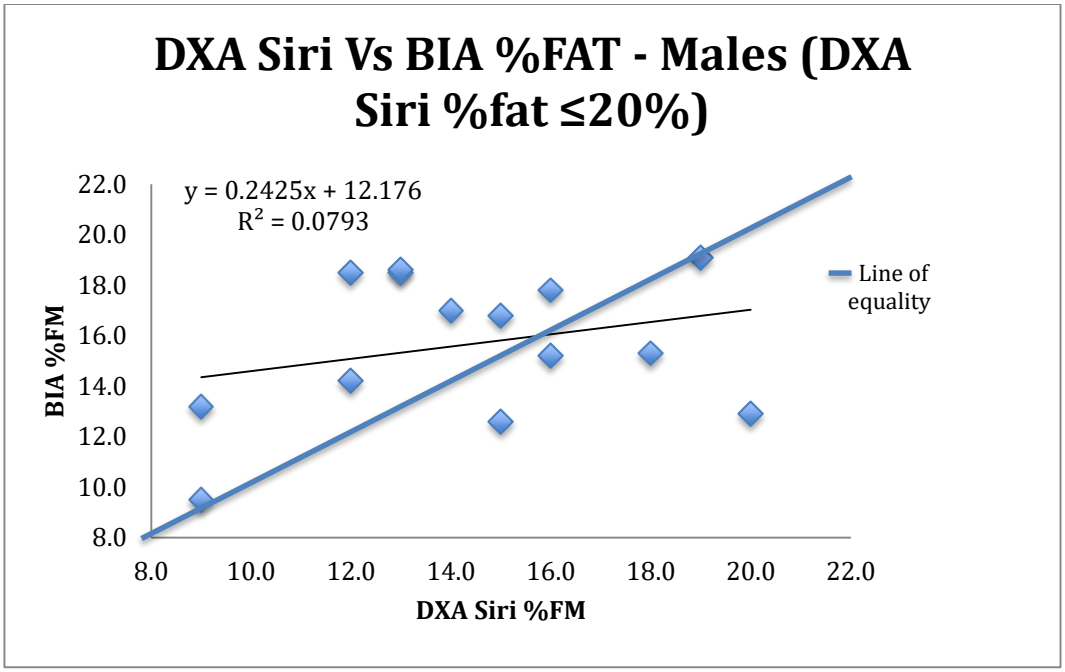
At the higher adiposity cut-offs, significant differences were observed between BIA and DXA_{Siri} %FM for both genders, with BIA consistently underestimating %FM (mean difference = 6.03%, P< 0.001 for boys; and mean difference = 5.73%, p = 0.004 for girls). However, for the male sample the correlation (Table 4.3 and Figure 4.22) was strong (r = 0.73, R^2 = 0.56) unlike their leaner counterparts, whilst for the female sample the correlation remained strong (r = 0.58, R^2 = 0.33).

These analyses were repeated for both genders at different age cut-offs. Male age cut-offs were ≤12y and > 12y, and female age cut-offs were ≤13y and >13y. Scatterplots for both age cut-offs are presented in Figures 4.23 to 4.24 and results for paired-samples tests are presented in Table 4.3. For both genders, correlations were strong for both age cut-offs, although the strongest correlations were observed

in the younger age groups (r = 0.82, R^2 = 0.68 for boys ≤12y; r = 0.85, R^2 = 0.72 for girls ≤13y; and r = 0.53, R^2 =0.28 for boys >12y; and r = 0.80, R^2 = 0.64 for girls > 13y). Significant differences were observed between BIA and DXA_{Siri} %FM for boys ≤12y, with BIA on average underestimating %FM (mean difference = 3.84%, p = 0.013. However, no significant differences were observed in boys >12y (mean difference = 0.49%, p = 0.732. In the female sample, differences were very close to significant in the younger age group (≤13y; mean difference = 3.51%, p = 0.055). In the older age group (>13y), no significant differences were observed (mean difference = 2.08%, p = 0.168).

Comparisons between BIA and DXA_{Siri} for absolute FM and FFM values as expected gave equivalent outputs but in the opposite direction. For this reason, only FM data is presented. Correlations were very strong in all cases (Table 4.3). For DXA_{Siri} %FM cut-offs, paired-sample tests for FM were very similar to %FM outcomes (Table 4.3). No significant differences were observed between BIA FM (kg) and DXA_{Siri} FM2 (kg) in the leaner samples for both genders (mean difference = -0.56kg & 0.21kg; sd = 2.2 & 1.73; p = 0.361 & 0.667; for boys and girls respectively). For the samples with higher adiposity, significant differences were observed, with BIA consistently underestimating FM in both genders (mean difference = 2.06kg & 2.33kg; sd = 1.18 & 2.05; P< 0.001 & = 0.004 for boys and girls respectively).

Comparisons between DXA_{Siri} and BIA FM with age cut-offs also gave very similar outcomes to %FM results. All variables were strongly correlated (Table 4.3). However, significant differences were observed between DXA_{Siri} FM2 and BIA FM for the younger age groups in both genders (mean difference = 1.11kg & 1.25kg; sd = 1.47 & 1.75; p = 0.014 & 0.03 for boys and girls respectively). No significant differences were observed for the older age group in both genders (mean difference = 0.26kg & 1.11kg; sd = 2.78 & .54; p = 0.737 & 0.157 for boys and girls respectively). In summary, in both genders there were no significant differences between BIA and DXA_{Siri} variables for both %FM and absolute FM (kg) for the leaner and older samples, with significant differences in both genders with higher adiposity and younger age.



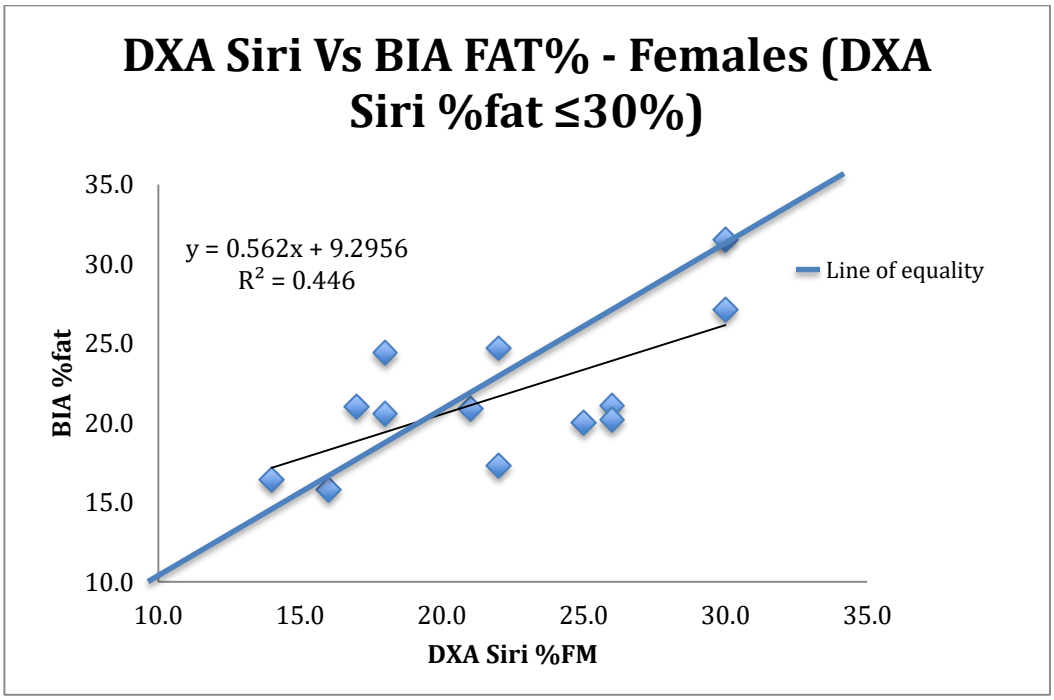
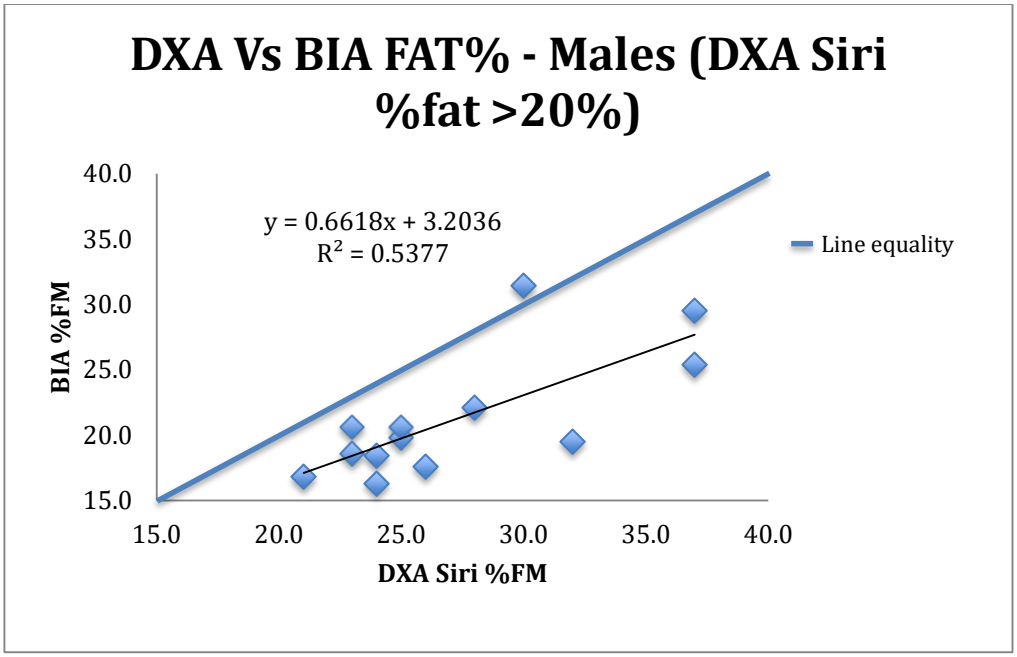


Figure 4.20 Scatterplots comparing DXASiri %FM with BIA %FM at ≤20% FM boys (upper graph) & ≤30% FM girls (lower graph). FM = fat mass; DXA = dual-energy x-ray absorptiometry; BIA= bioelectrical impedance analysis; DXAsiri (see section 4.3.3).



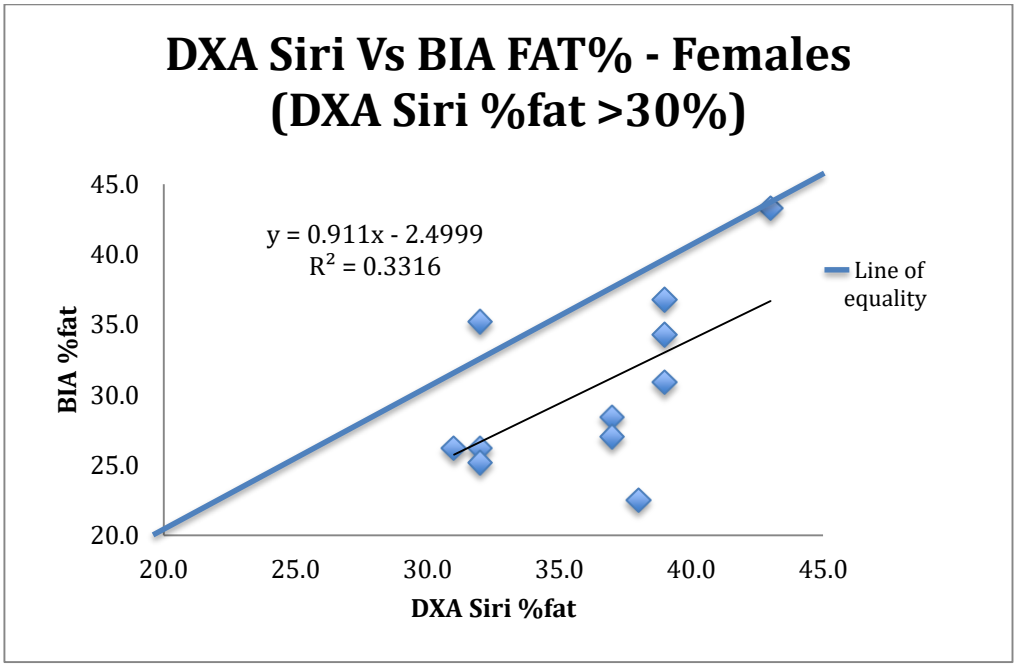
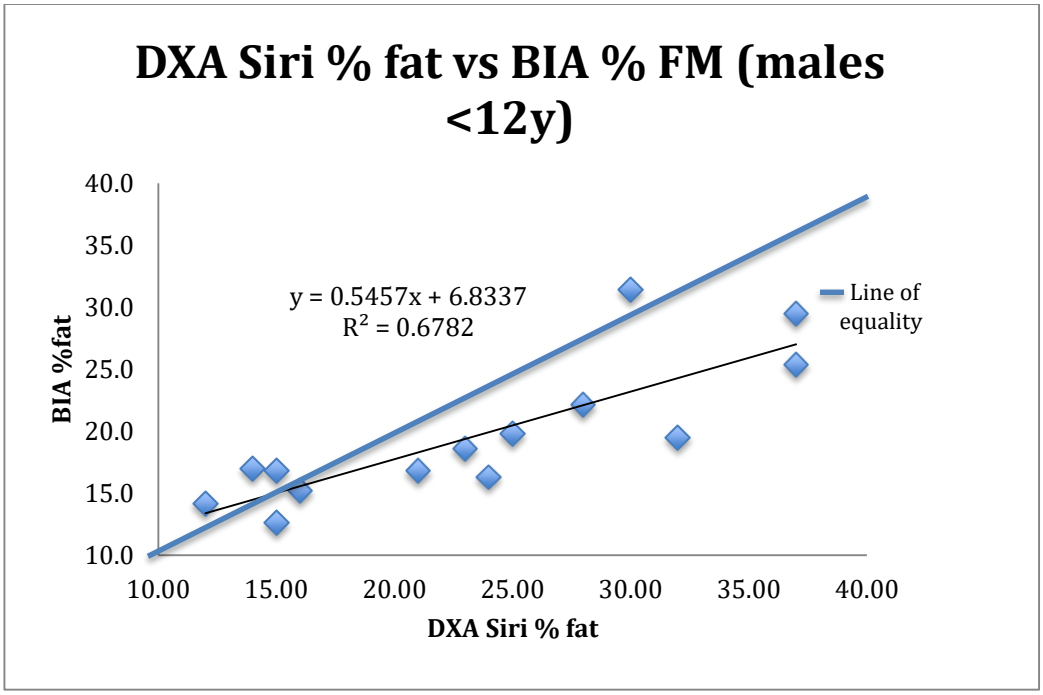


Figure 4.21 Scatterplots comparing DXASiri %FM with BIA %FM at >20% FM boys (upper graph) & >30% FM girls (lower graph). FM = fat mass; DXA = dual-energy x-ray absorptiometry; BIA= bioelectrical impedance analysis; DXAsiri (see section 4.3.3).



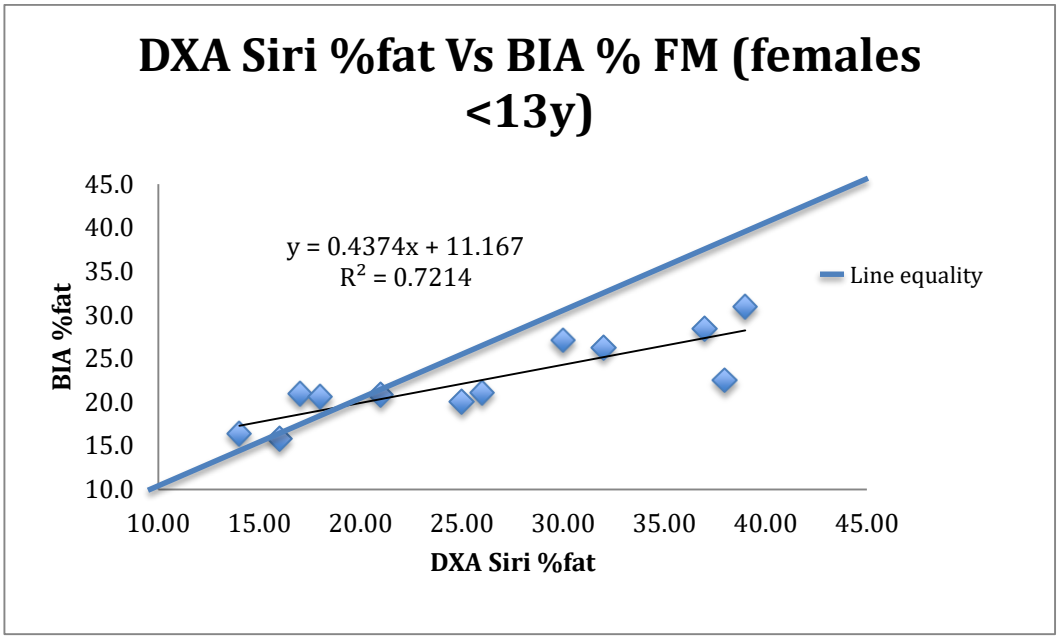
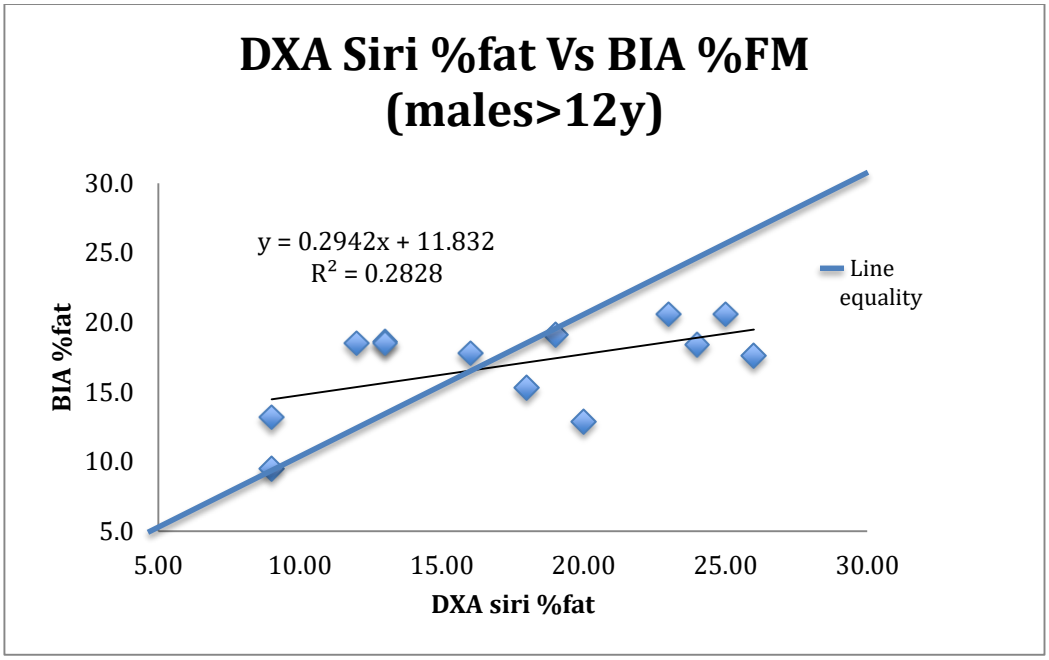


Figure 4.22 Scatterplots comparing DXASiri %FM with BIA %FM at ≤12y boys (upper graph) & ≤13y girls (lower graph). FM = fat mass; DXA = dual-energy x-ray absorptiometry; BIA= bioelectrical impedance analysis; DXAsiri (see section 4.3.3).



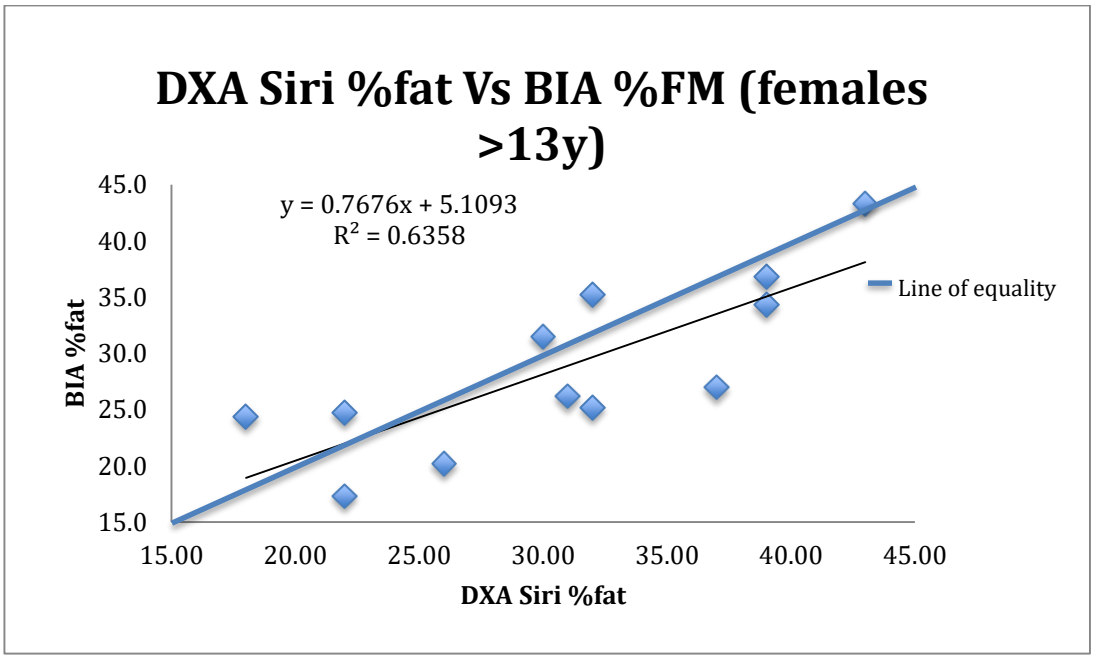


Figure 4.23 Scatterplots comparing DXASiri %FM with BIA %FM at >12y boys (upper graph) & >13y girls (lower graph). FM = fat mass; DXA = dual-energy x-ray absorptiometry; BIA= bioelectrical impedance analysis; DXAsiri (see section 4.3.3).

Table 4.3: Body composition (mean ± sd) measures of SA BIA validation study group by BIA and DXA, for percentage fat-mass (%FM), absolute fat-mass (FM; kg), with Bland-Altman comparisons between BIA and reference method showing mean difference and limits of agreement (mean ± 2sd)

Boys					**Girls**			
	N	**Mean ± sd**	**Mean difference ± sd, & correlation (r) DXA_{Siri} - BIA**	**Limits of agreement (mean difference ± 2sd) between BIA & DXA_{Siri}**	**N**	**Mean ± sd**	**Mean difference ± sd, & correlation (r) DXA_{Siri} - BIA**	**Limits of agreement (mean difference ± 2sd) between BIA & DXA_{Siri}**
BIA FMℜ (%)	14	15.66 ± 2.88	-1.30 ± 3.75, (0.28)		13	21.62 ± 4.39	-0.31 ± 3.99, (0.67)	
DXA_{Siri} FMℜ (%)	14	14.36 ± 3.34	-		13	21.92 ± 5.22	-	
BIA FMℑ (%)	13	21.28 ± 4.72	6.03 ± 3.67, (7.33) **		11	30.55 ± 6.21	5.73 ± 5.09, (0.58) *	
DXA_{Siri} FMℑ (%)	13	27.31 ± 5.23	-		11	36.27 ± 3.93	-	
BIA FMδ (%)	14	19.66 ± 5.61	3.84 ± 4.99, (0.82) *		12	22.58 ± 4.66	3.51 ± 5.65, (0.85)	
DXA_{Siri} FMδ (%)	14	23.50 ± 8.46	-		12	26.08 ± 9.05	-	
BIA FMχ (%)	13	16.97 ± 3.30	0.49 ± 5.05, (0.53)	-9.62 to 10.60%	12	28.84 ± 7.50	2.08 ± 4.87, (0.80)	
DXA_{Siri} FMχ (%)	13	17.46 ± 5.97	-		12	30.92 ± 7.79	-	
BIA FMℜ (kg)	14	7.10 ± 3.85	-0.56 ± 2.20, (0.84)		13	8.98 ± 4.66	0.21 ± 1.73, (0.93)	
DXA_{Siri} FM2ℜ (kg)	14	6.54 ± 3.99	-		13	9.19 ± 4.75	-	
BIA FMℑ (kg)	13	8.32 ± 4.53	2.06 ± 1.18, (0.97) **		11	16.61 ± 9.09	2.33 ± 2.05, (0.98) *	
DXA_{Siri} FM2ℑ (kg)	13	10.38 ± 4.72	-		11	18.94 ± 7.99	-	
BIA FMδ (kg)	14	5.71 ± 3.23	1.11 ± 1.47, (0.93) *		12	7.52 ± 3.31	1.25 ± 1.75, (0.95) *	
DXA_{Siri} FM2δ (kg)	14	6.82 ± 3.83	-		12	8.77 ± 4.53	-	
BIA FMχ (kg)	13	9.81 ± 4.08	0.26 ± 2.78, (0.84)		12	17.43 ± 8.11	1.11 ± 2.54, (0.95)	-3.96 to 6.18kg
DXA_{Siri} FM2χ (kg)	13	10.07 ± 5.10	-		12	18.54 ± 7.84	-	

SA = South Asian; BIA = Bioelectrical Impedance Analysis; DXA = Dual-energy x-ray absorptiometry; ℜ DXA_{Siri} %FM cut-offs ≤ 20% and ≤ 30%; ℑ DXA_{Siri} %FM cut-offs > 20% and > 30% for boys and girls respectively.δ Age cut-offs < 12y and < 13y; χ age cut-offs > 12y and > 13y for boys and girls respectively. *P< 0.05; **P< 0.001 Sig. (2–tailed). DXA_{Siri} FM2 (kg), was weight corrected using BIA scale weight (see section 4.5.2).

4.5.3.5 Linear Regression Analyses to Predict FFM

Whilst the comparisons between BIA and DXA_{Siri} %FM for each gender at different age and %FM cut-offs yielded some outputs with non-significant differences, splitting the subjects in this manner resulted in very small sample sizes. Thus, to draw any meaningful conclusions from these results would necessitate repeating the analyses with a much larger sample size.

Further analyses were conducted using standard linear regression, to establish which BIA predictor variables were the most significant predictors of FFM, using DXA_{Siri} FFM2 (Wt corrected) as the dependent variable (DV), and age, gender, Wt, Ht, BMI, WC, and Ht^2/Z as the independent predictors.

With all variables entered into the model the adjusted R^2 value = 0.982, indicating that 98.2% of the variance in DXA Siri FFM2, was explained by this model. For both genders the largest (Beta values) and significant ($P< 0.05$) predictors of DXA Siri FFM2, were Ht^2/Z ($P<0.001$), followed by BIA Wt ($p = 0.008$), and decimal age ($p = 0.046$); gender was not a significant predictor of FFM. Significant bivariate correlations ($R \geq 0.7$) were observed between BIA Wt, Ht, BMI, WC, and Ht^2/Z, indicating multicollinearity, with tolerance values of < 0.1 and variance inflation factor (VIF) values of >10, thus violating the multicollinearity assumption (Pallant, 2007).

Regression analyses were repeated with Ht^2/Z, BIA Wt, and decimal age only. With this model, the adjusted R^2 value = 0.973, which compared to the value for the previous model, confirmed that the other variables were not making a significantly unique contribution to the prediction model (Table 4.4). The ANOVA output confirmed the model to be statistically significant ($P< 0.001$).

Analyses of the normal probability plot (P-P) of the regression standardised residual (Figure 4.24) and the scatterplot (Figure 4.25) indicated that there was a linear relationship between DXA_{Siri} FFM2 (DV) and the prediction model, with no major deviations from normality or outliers.

Table 4.4: SPSS output - Model summary[b] showing the relationship (r & r^2) between the BIA prediction model for predicting FFM (kg) and DXA_{Siri} FFM2 using linear regression

Model		r	r square	Adjusted r square	Std. Error of Estimate
1		0.987[a]	0.975	0.973	2.21

a. Predictors: (Constant), Decimal Age (yrs), BIA Wt (kg), Ht^2/impedance – Wt = weight; Ht= height; BIA = Bioelectrical impedance analysis

Dependent variable: DXA_{Siri} FFM2 (kg), were weight corrected using BIA scale weight (see section 4.5.2).

b. FFM = fat-free mass

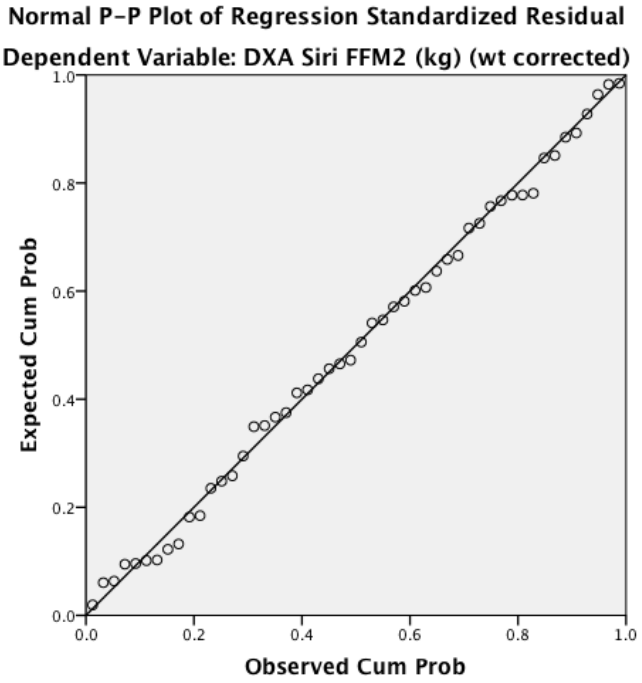


Figure 4.24 SPSS output showing normal P-P plot of regression standardised residual with DXASiri FFM2 as dependent variable. FFM = fat-free mass; DXA= dual-energy x-ray absorptiometry; DXASiri FFM2 = DXASiri FFM2 weight (Wt) corrected (see section 4.5.2).

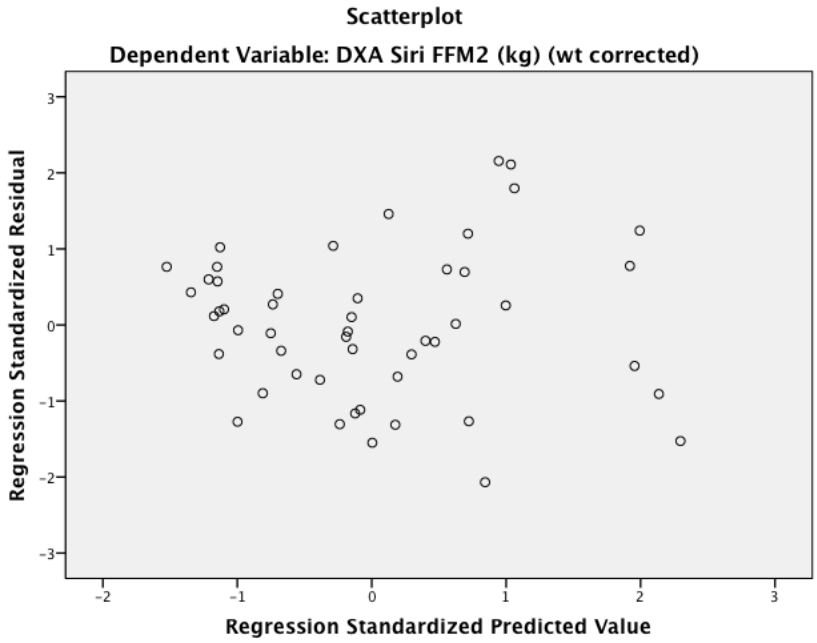


Figure 4.25 Scatterplot of regression standardised predicted value Vs regression standardised residual with DXASiri FFM2 as dependent variable. FFM = fat-free mass; DXA= dual-energy x-ray absorptiometry; DXASiri FFM2 = DXASiri FFM2 weight (Wt) corrected (see section 4.5.2).

The regression equation developed using the unstandardized coefficients labelled B, from the SPSS 'Coefficients' output tables (see Table 4.5). The regression equation to predict BIA FFM (BIA $FFM_{reg1)}$ was determined as follows:

BIA FFM_{reg1} (kg) = (0.834 x Ht^2/Z) + (0.182 x BIA Wt) + (0.322 x Decimal Age) – 4.862

Table 4.5: SPSS output - Unstandardised Coefficients[a] for determination of BIA_{reg1} prediction equation with DXA_{Siri} FFM2 as dependent variable

Model 1 Predictors	**Unstandardised Coefficients B**
(Constant)	-4.862
Ht^2/impedance	0.834
BIA Wt (kg)	0.182
Decimal Age (yrs)	0.322

Ht = height; Wt = Weight; FFM = fat-free mass; BIA = Bioelectrical impedance analysis = BIA_{reg1} = BIA regression 1 prediction equation (see section 4.5.3.5)

a. Dependent variable: DXA_{Siri} FFM2 (kg), was weight corrected using BIA scale weight (see section 4.5.2).

4.5.3.6 Statistical comparisons between DXA_{Siri} and BIA for FFM, %FM and absolute FM, following regression

Due to body composition differences between genders, the new regression equation was assessed for each sex separately. As the equation predicted FFM (BIA FFM_{reg1}), FM (kg) and %FM values could also be determined, where BIA FM_{reg1} (kg) = BIA Wt – BIA FFM_{reg1}, and BIA $\%FM_{reg1}$ = (BIA FM_{reg1}/BIA Wt) x 100. The relationship and differences between DXA_{Siri} and BIA_{reg1} for these body composition variables were compared, using correlations and t-tests (Table 4.6). Outcomes for the relationship and difference between BIA and DXA_{Siri} prior to correction are also presented, for ease of comparison.

In Table 4.6, correcting BIA FFM using the regression equations decreased the mean differences between BIA and DXA for both sexes. The significance values also improved, although for the male sample this was not significantly different before correction. For boys, before correction, compared to DXA_{Siri} FFM_2, BIA overestimated FFM by a mean of 0.56kg (sd = 2.24; limits of agreement = -5.10kg to 3.70kg; p = 0.202); after correction, BIA reg1 underestimated FFM by a mean of 0.47kg (sd = 2.15; limits of agreement = -3.73kg to 4.75kg; p = 0.267). For girls, before correction, BIA was significantly overestimating FFM by a mean of 1.25kg (sd = 2.16, limits of agreement = -5.45kg to 3.08kg, p = 0.011); after correction, the difference dropped, with BIA FFM reg1 overestimating FFM by a mean of 0.51kg (sd = 2.04, limits of agreement = -4.61kg to 3.55kg, p = 0.225).

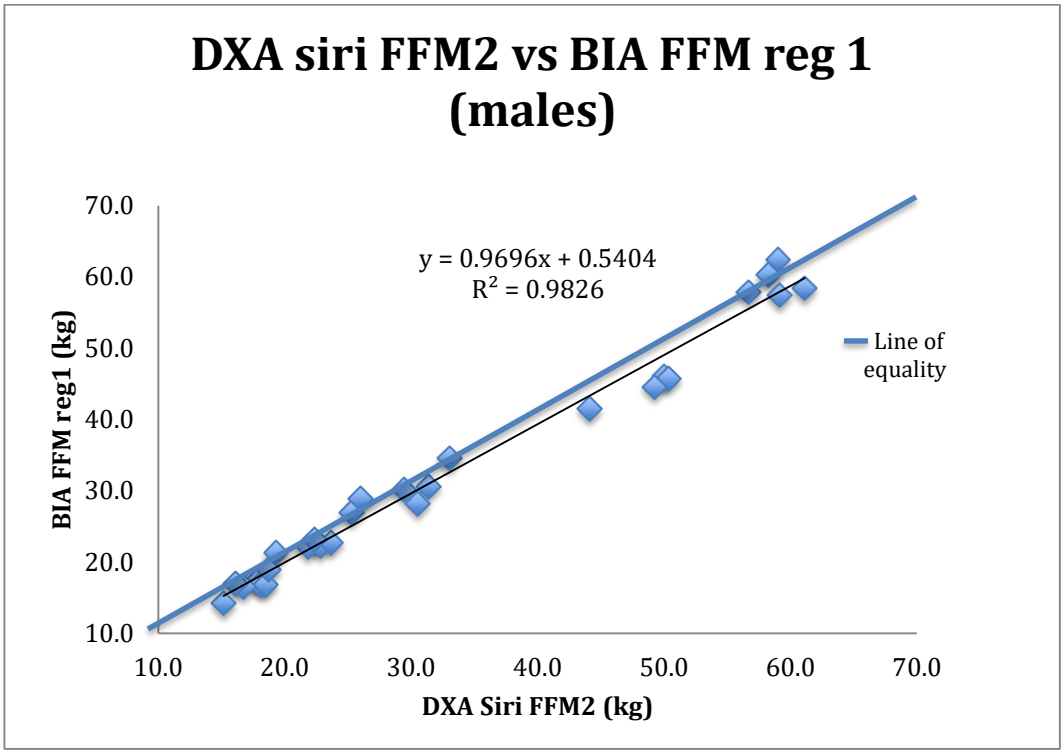
Before correction, BIA was underestimating %FM in both sexes. However, after correction the differences in both sexes were non-significant. For boys, before correction, BIA was underestimating %FM by a mean of 2.23% (sd = 5.21, limits of agreement = -8.19% to 12.65%, p = 0.035); after correction BIA_{reg1} was overestimating %FM by 1.07% (sd = 4.71, limits of agreement = -10.48% to 8.34%, p = 0.248). For girls, before correction, BIA was underestimating %FM by a mean of 2.79% (sd = 5.21, limits of agreement = -7.65% to 13.50%, p = 0.015); after correction BIA_{reg1} was underestimating %FM by 0.73% (sd = 5.74, limits of agreement = -10.75% to 12.20%, p = 0.549).

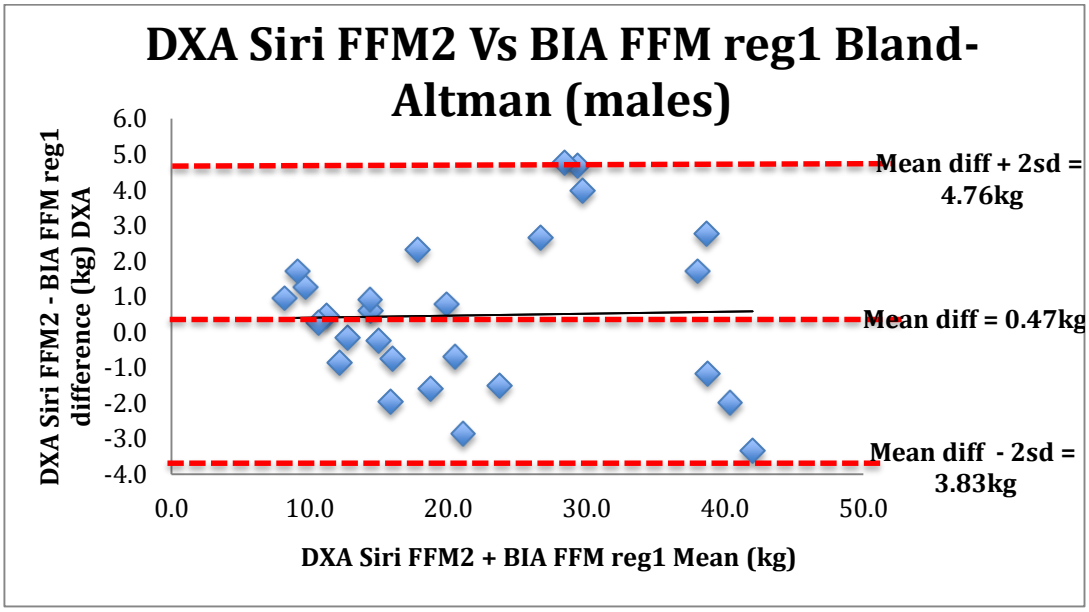
Scatterplots and Bland-Altman plots by gender, showing the correlations and mean differences respectively, between BIA FFM_{reg1} and DXA_{Siri} FFM2, and BIA $\%FM_{reg1}$ and DXA_{Siri} %FM are presented in Figures 4.27 to 4.28. The scatterplots and Bland-Altman plots for FFM (Figure 4.26), show how well BIA FFM_{reg1} correlates with DXA_{Siri} FFM2, however, the scatterplots for %FM (Figure 4.27), indicate that BIA $\%FM_{reg1}$ has a tendency to overestimate %FM at the leaner end of the scale (≤ 15%FM in boys and ≤ 20% FM in girls), and underestimate %FM at higher adiposity levels. This outcome was similar to the original BIA %FM outcome before correction (see section 4.5.3.1), although the mean difference was smaller and non-significant following correction.

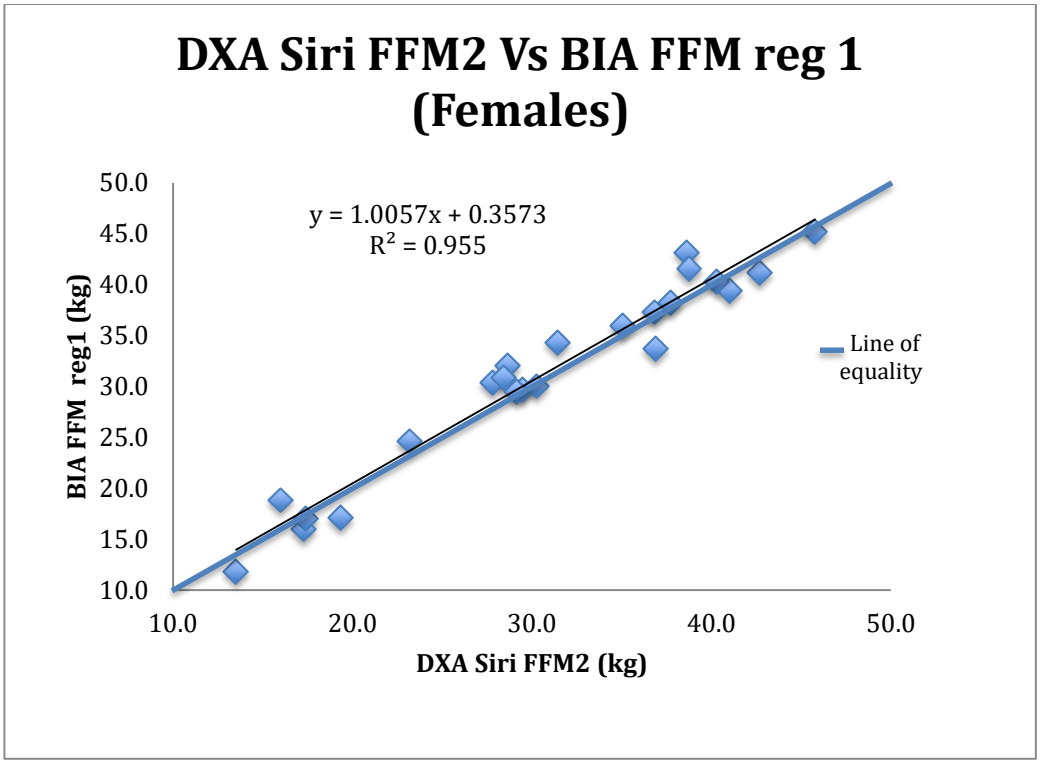
Table 4.6 Body composition measures (mean ±sd) of SA BIA validation study group by BIA and DXA_{Siri}, for relative fat mass (%; FM), and absolute fat-free mass (kg; FFM), with comparisons showing mean differences, correlations, and Bland-Altman limits of agreement, between methods, before and after BIA correction

Variables	**Boys (n = 27)**			**Girls (n = 24)**		
	Mean ± sd	**Mean difference ± sd, (r) DXA_{Siri} - BIA**	**Limits of agreement (mean difference ± 2sd) DXA_{Siri} - BIA**	**Mean ± sd**	**Mean difference ± sd, (r) DXA_{Siri} - BIA**	**Limits of agreement (mean difference ± 2sd) DXA_{Siri} - BIA**
DXA_{Siri} FFM2¶ (kg)	33.16 ± 16.18	-	-	30.68 ± 9.34	-	-
BIA FFM_{reg1} (kg)	32.69 ± 15.83	0.47 ± 2.15, (0.99)	-3.73 to 4.75	31.22 ± 9.61	-0.51 ± 2.04, (0.98)	-4.61 to 3.55
BIA FFM (kg)	33.72 ±15.71	-0.56 ± 2.24, (0.99)	-5.10 to 3.70	31.93 ± 9.30	-1.25 ± 2.16, (0.97) *	-5.45 to 3.08
DXA_{Siri} FM (%)	20.59 ± 7.86	-	-	27.87 ± 8.23	-	-
BIA FM_{reg1} (%)	21.66 ± 6.28	-1.07 ± 4.71, (0.80)	-10.48 to 8.34	27.14 ± 6.42	0.73 ± 5.74, (0.72)	-10.75 to 12.20
BIA FM (%)	18.54 ± 4.92	2.23 ± 5.21, (0.77) *	-8.19 to 12.65	24.94 ± 5.91	2.79 ± 5.79, (0.80) *	-7.65 to 13.50

SA = South Asian; DXA = Dual-energy x-ray absorptiometry; BIA = Bioelectrical impedance analysis; BIA FFM_{reg1} & FM_{reg1} is corrected following new equation derived from regression between BIA and DXA_{Siri} FFM2 (see section 4.5.3.5)
* P< 0.05 Sig. (2-tailed)
¶DXA_{Siri} FFM2 (kg) is Wt corrected using BIA scale Wt (see section 4.5.2)


DXA siri FFM2 vs BIA FFM reg 1 (males)
y = 0.9696x + 0.5404
R² = 0.9826
Line of equality
BIA FFM reg1 (kg)
DXA Siri FFM2 (kg)
70.0
60.0
50.0
40.0
30.0
20.0
10.0
10.0
20.0
30.0
40.0
50.0
60.0
70.0


DXA Siri FFM2 Vs BIA FFM reg1 Bland-Altman (males)
DXA Siri FFM2 - BIA FFM reg1 difference (kg) DXA
DXA Siri FFM2 + BIA FFM reg1 Mean (kg)
Mean diff + 2sd = 4.76kg
Mean diff = 0.47kg
Mean diff - 2sd = 3.83kg
6.0
5.0
4.0
3.0
2.0
1.0
0.0
-1.0
-2.0
-3.0
-4.0
0.0
10.0
20.0
30.0
40.0
50.0



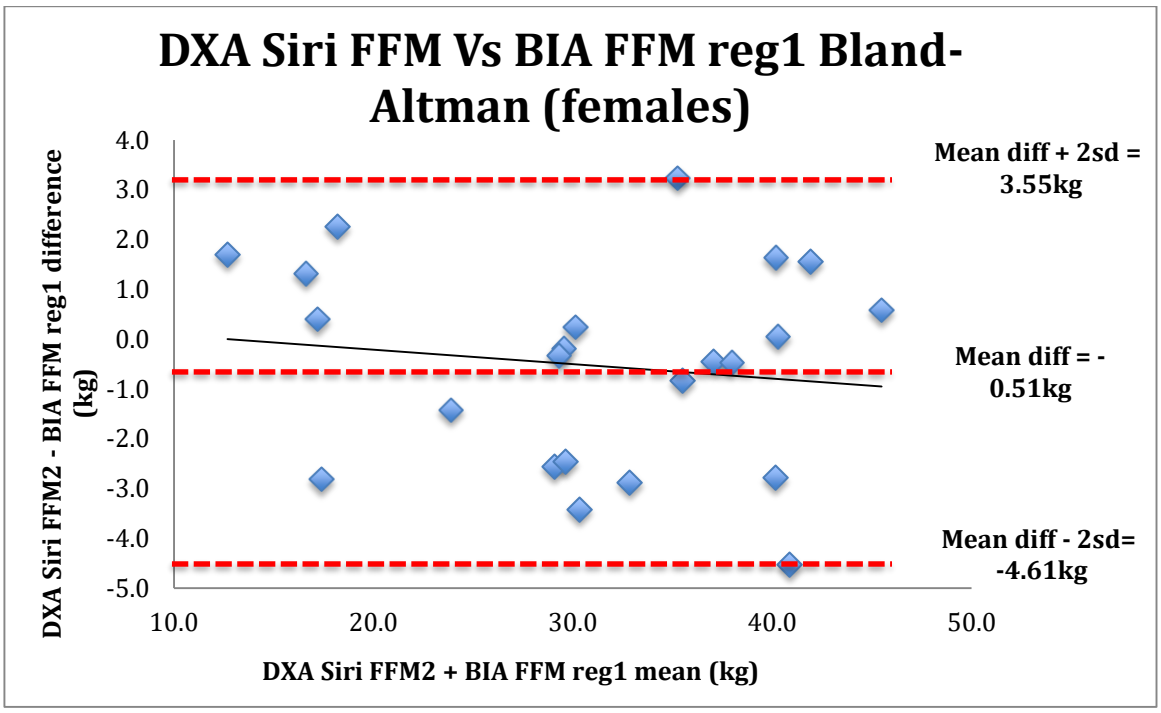
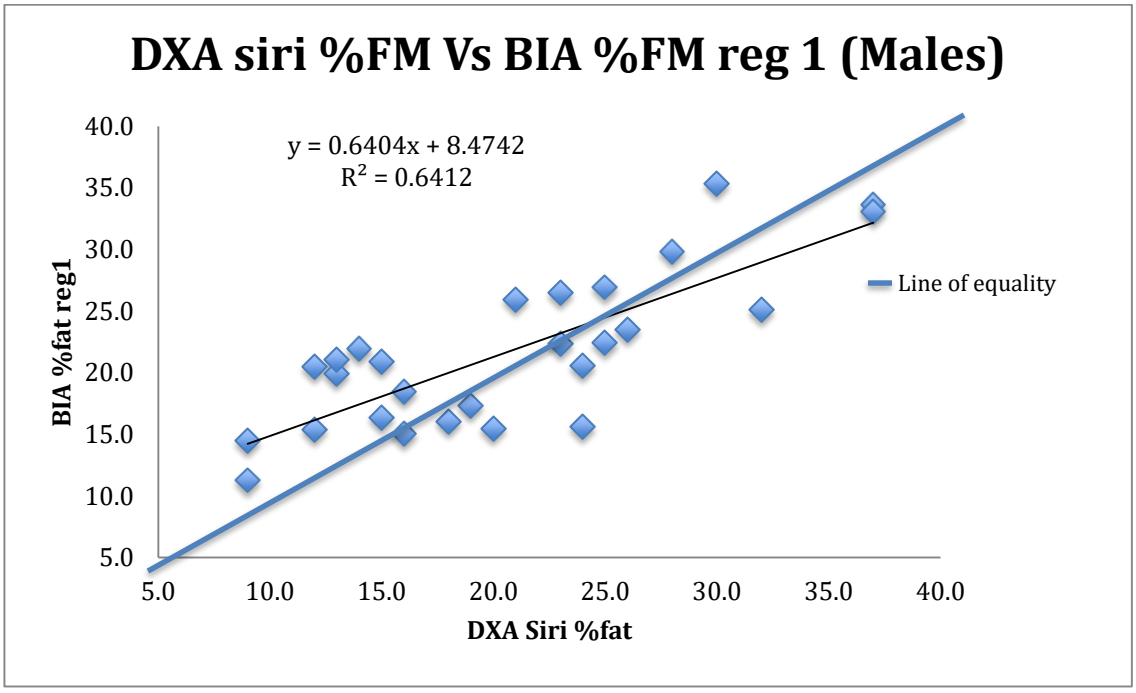
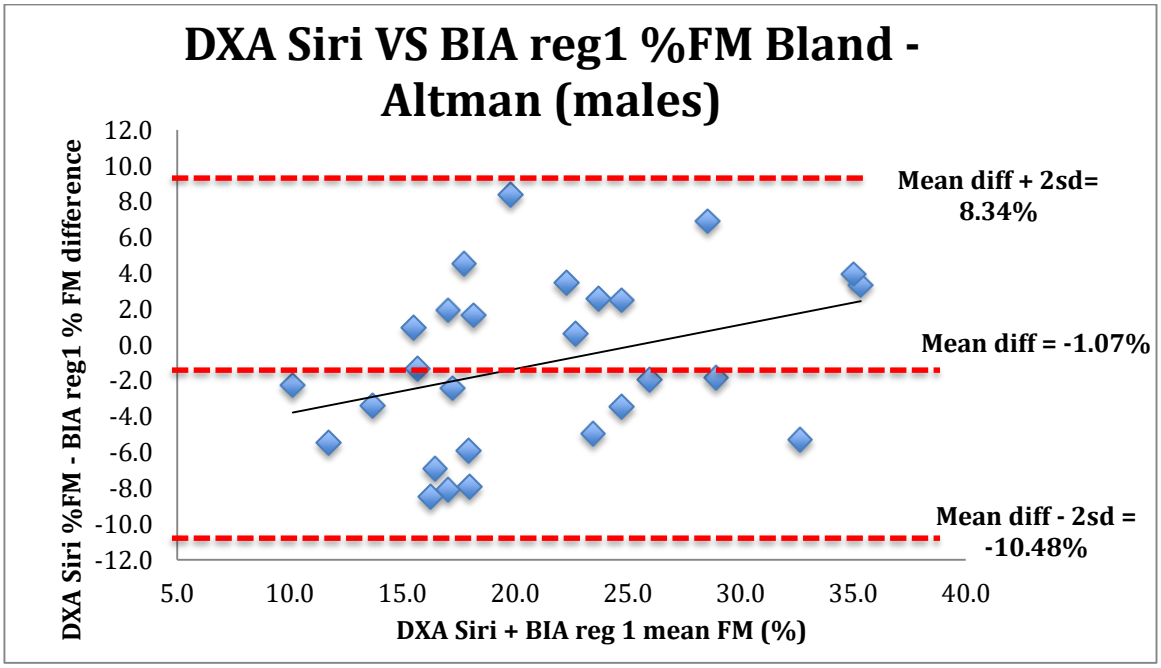
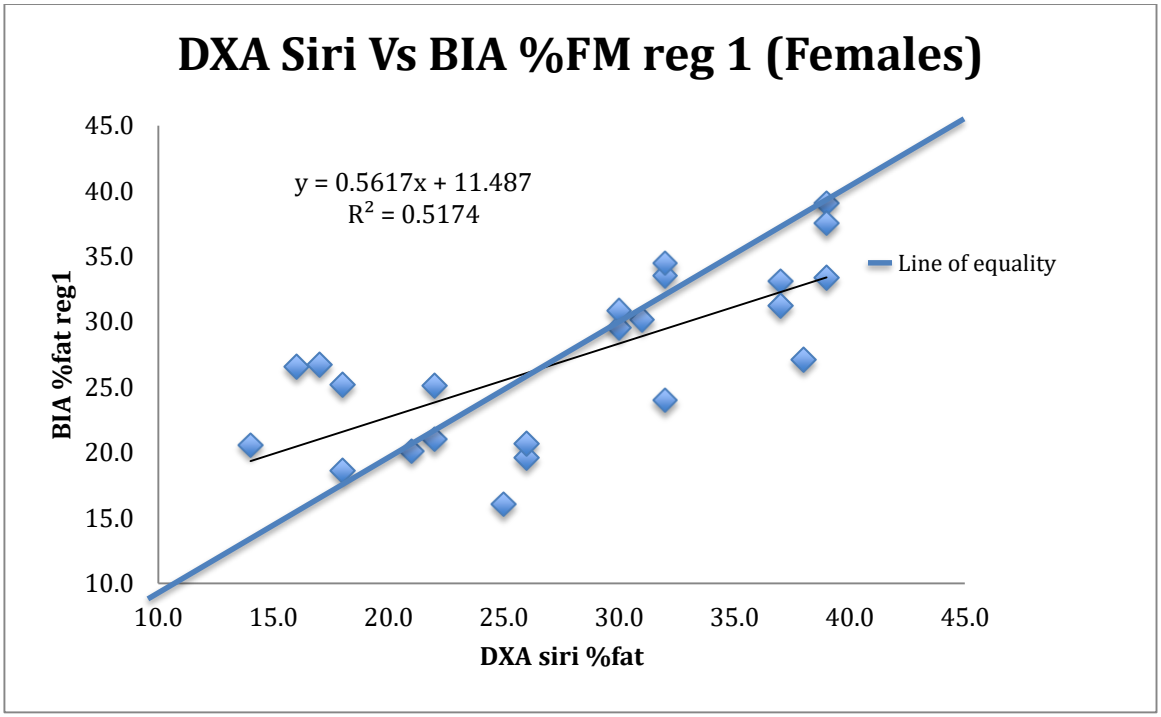


Figure 4.26 Scatterplots showing correlations (upper graph), and Bland-Altman plots showing limits of agreement (lower graph) by gender, between DXASiri FFM2 and BIA FFMreg1; BIA FFMreg1 = new BIA FFM following regression (see section 4.5.3.5); DXASiri FFM2 = DXA weight corrected (see section 4.5.2). FFM = fat-free mass; DXA = dual-energy x-ray absorptiometry; BIA = bioelectrical impedance analysis.


DXA siri %FM Vs BIA %FM reg 1 (Males)
y = 0.6404x + 8.4742
R² = 0.6412
Line of equality
BIA %fat reg1
DXA Siri %fat
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
5.0
10.0
15.0
20.0
25.0
30.0
35.0
40.0


DXA Siri VS BIA reg1 %FM Bland - Altman (males)
Mean diff + 2sd= 8.34%
Mean diff = -1.07%
Mean diff - 2sd = -10.48%
DXA Siri %FM - BIA reg1 % FM difference
DXA Siri + BIA reg 1 mean FM (%)
12.0
10.0
8.0
6.0
4.0
2.0
0.0
-2.0
-4.0
-6.0
-8.0
-10.0
-12.0
5.0
10.0
15.0
20.0
25.0
30.0
35.0
40.0



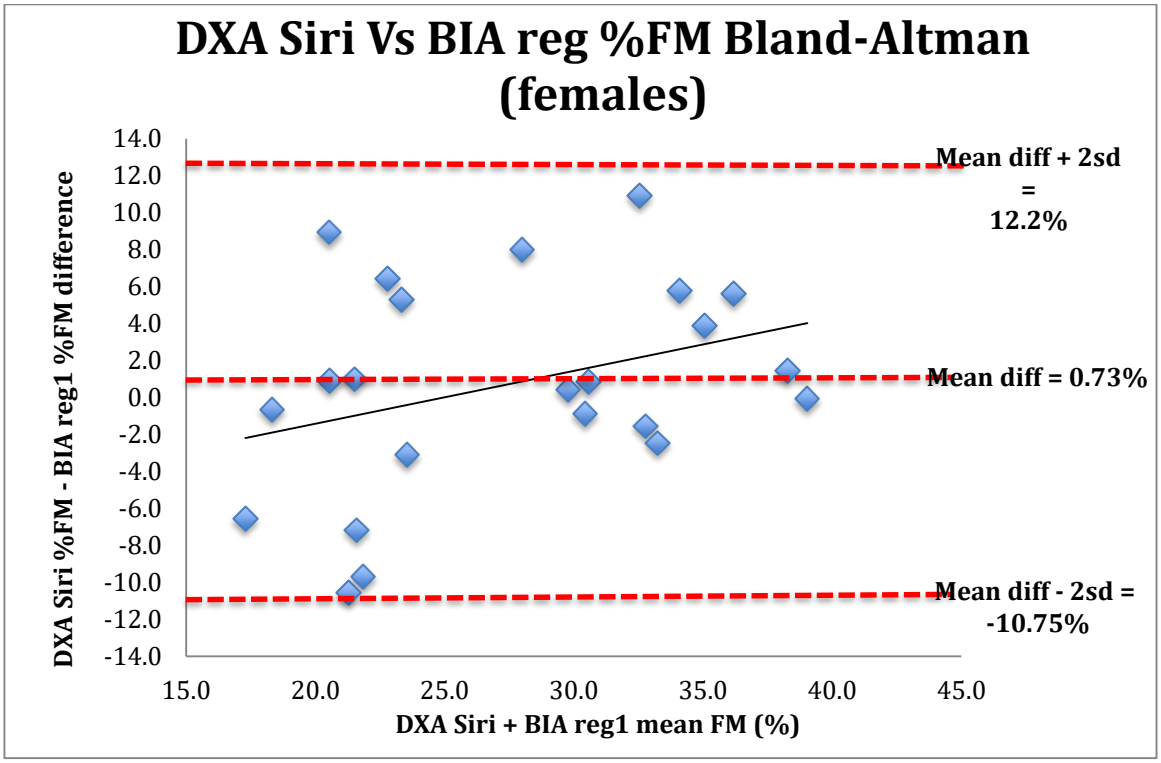


Figure 4.27 Scatterplots showing correlations (upper graph), and Bland-Altman plots showing limits of agreement (lower graph) by gender, between DXASiri %FM and BIA %FMreg1. FM = fat mass; BIA FMreg1 = new BIA FM following regression (see section 4.5.3.5); DXA = dual-energy x-ray absorptiometry; BIA = bioelectrical impedance analysis.

4.5.3.7 Application of BIA correction factor to SA external sample

The correction factors obtained from the regression of BIA FFM with DXA_{Siri} FFM2 (section 4.5.3.5) were applied to the SA external sample, to assess the differences in BIA absolute (kg) FFM and relative FM (%), before and after correction. Initial observation of the results for %FM between BIA and BIA_{reg1} revealed that for both genders, BIA_{reg1} produced much higher %FM values for children <9y, which were considered spurious. As the study group was significantly older than the external group (Table 4.1), with very few children below 9y, more detailed comparisons were restricted to children aged 9y and above. Significant differences ($P<0.001$) were observed between all measures for both absolute and relative FFM and FM, in both sexes between BIA (uncorrected) and BIA_{reg1} (Table 4.7). Scatterplots showing the relationship and mean differences (Bland-Altman plots) between BIA and BIA_{reg1}, for both absolute FFM (kg) and relative FM (%) are presented in Figures 4.28 and 4.29 respectively.

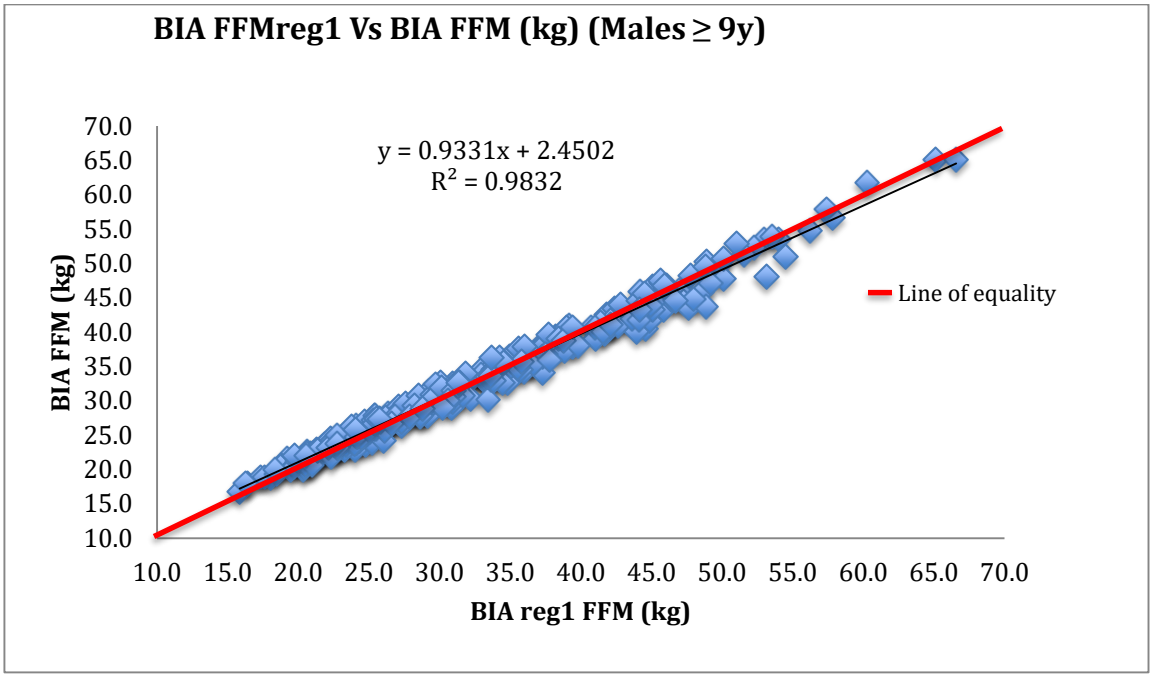
There was a strong positive correlation between BIA and BIA_{reg1} ($R^2= 0.98$) in FFM for both sexes (Figure 4.28). The Bland-Altman plots for FFM revealed that compared to $BIA_{reg1,}$ BIA overestimated FFM by a mean of 0.33kg (limits of agreement -2.99kg to 2.53kg) in boys, and by 0.27kg (limits of agreement -2.29kg to 1.74kg) in girls. For boys, the Bland-Altman plot also revealed that compared to BIA_{reg1}, BIA had a tendency to overestimate FFM at lower FFM levels, and underestimate FFM at higher FFM levels (Figure 4.28), which was similar for girls but less pronounced. For %FM (Figure 4.29), the results revealed that compared to BIA_{reg1}, BIA underestimated %FM by a mean of 1.10% (limits of agreement -5.51% to 7.71%) for boys, and by 0.67% (limits of agreement -4.70% to 6.04%). The scatterplots (Figure 4.29) also revealed that for both sexes, BIA_{reg1} compared to BIA tended to overestimate %FM at low (≤ 15%) body fat levels and underestimate %FM at higher body fat levels.

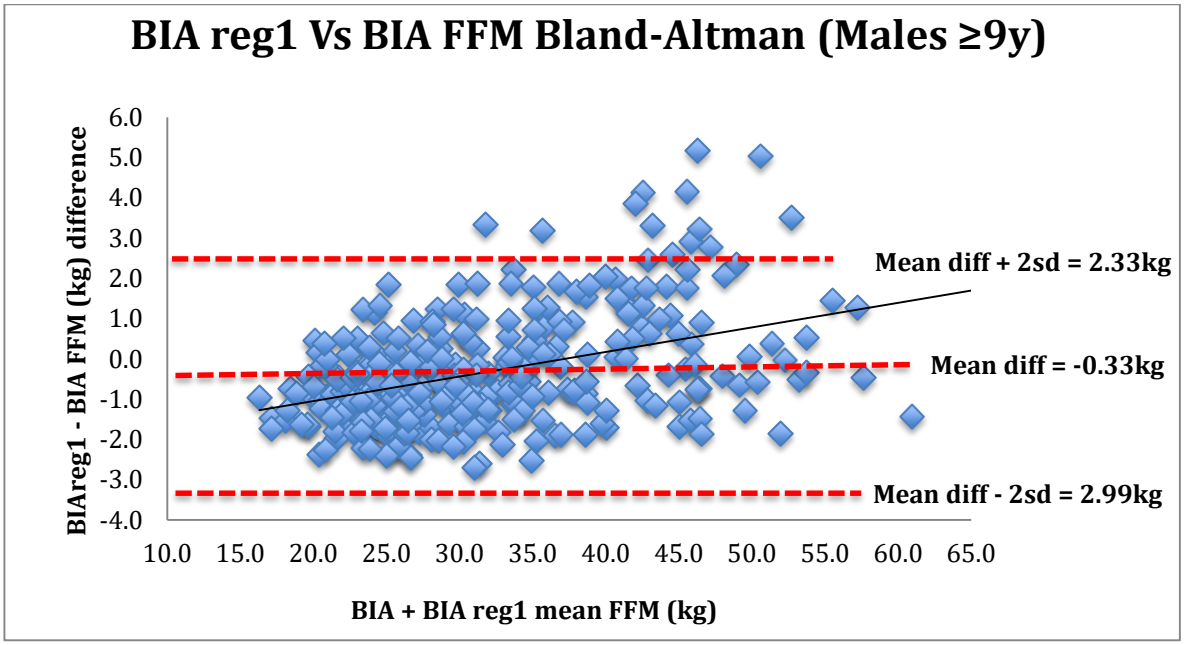
Table 4.7 Body composition measures (mean ±sd) of SA BIA validation study group by BIA and BIA_{reg1}, for absolute (kg) fat-free mass (FFM) and relative (%) FM, with comparisons showing mean differences, correlations, and Bland-Altman limits of agreement, between methods, before and after BIA correction

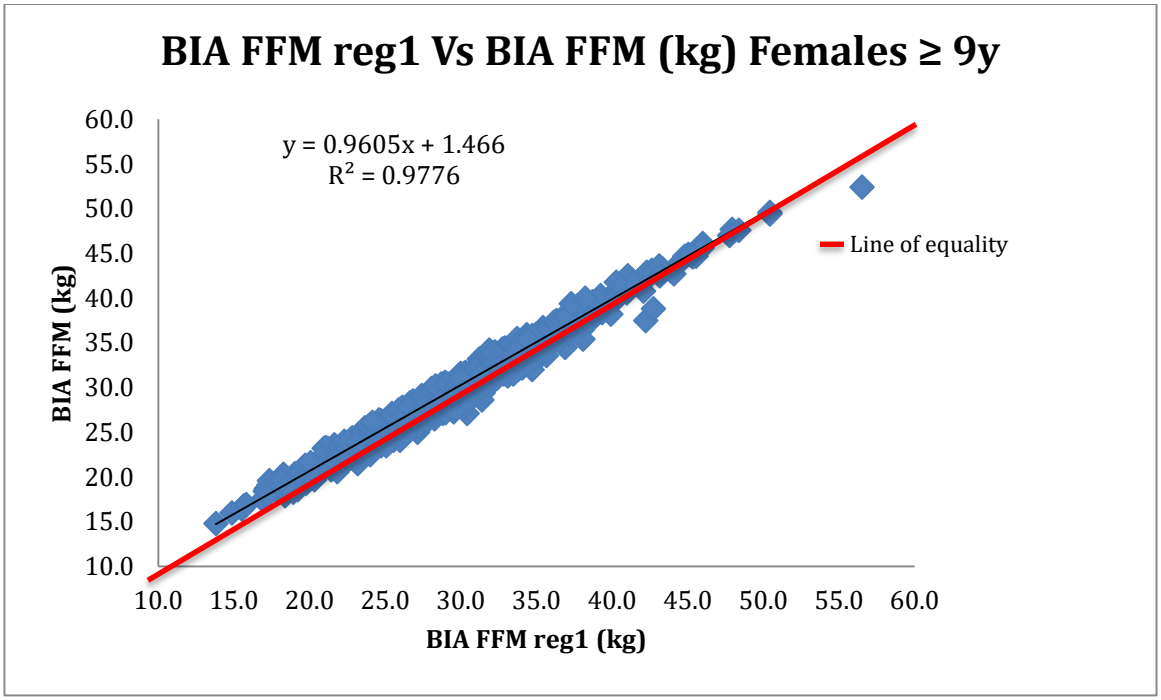
Variables	**Boys (n = 344)**			**Girls (n = 607)**		
	Mean ± sd	**Mean difference ±sd, (r) BIA_{reg1} - BIA**	**Limits of agreement (mean difference ± 2sd) BIA_{reg1} - BIA**	**Mean ±sd**	**Mean difference ± sd, (r) BIA_{reg1} - BIA**	**Limits of agreement (mean difference ±2sd) BIA_{reg1} - BIA**
BIA FFM_{reg1} (kg)	31.63 ± 8.80	-0.33 ± 1.33, (0.99) **	-2.99 to 2.33	30.25 ± 6.67	-0.27 ± 1.00, (0.99) **	-2.29 to 1.74
BIA FFM (kg)	31.96 ± 9.00			30.53 ± 6.48		
BIA FM_{reg1} (%)	22.30 ± 8.80	1.10 ± 3.30, (0.93) **	-5.51 to 7.71%	25.84 ± 7.60	0.67 ± 2.67, (0.95) **	-4.70 to 6.04
BIA FM (%)	21.20 ± 7.22			25.16 ± 6.16		

SA = South Asian; BIA = Bioelectrical impedance analysis; BIA FFM_{reg1} & FM_{reg1} is corrected following new equation derived from regression between BIA and DXA_{Siri} FFM2 (see section 4.5.3.5); DXA= Dual-energy x-ray absorptiometry

**P<0.001 Sig. (2-tailed)


BIA FFMreg1 Vs BIA FFM (kg) (Males ≥ 9y)
y = 0.9331x + 2.4502
R² = 0.9832
Line of equality
BIA FFM (kg)
BIA reg1 FFM (kg)
70.0
65.0
60.0
55.0
50.0
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
10.0 15.0 20.0 25.0 30.0 35.0 40.0 45.0 50.0 55.0 60.0 65.0 70.0


BIA reg1 Vs BIA FFM Bland-Altman (Males ≥9y)
BIAreg1 - BIA FFM (kg) difference
BIA + BIA reg1 mean FFM (kg)
Mean diff + 2sd = 2.33kg
Mean diff = -0.33kg
Mean diff - 2sd = 2.99kg
6.0
5.0
4.0
3.0
2.0
1.0
0.0
-1.0
-2.0
-3.0
-4.0
10.0 15.0 20.0 25.0 30.0 35.0 40.0 45.0 50.0 55.0 60.0 65.0



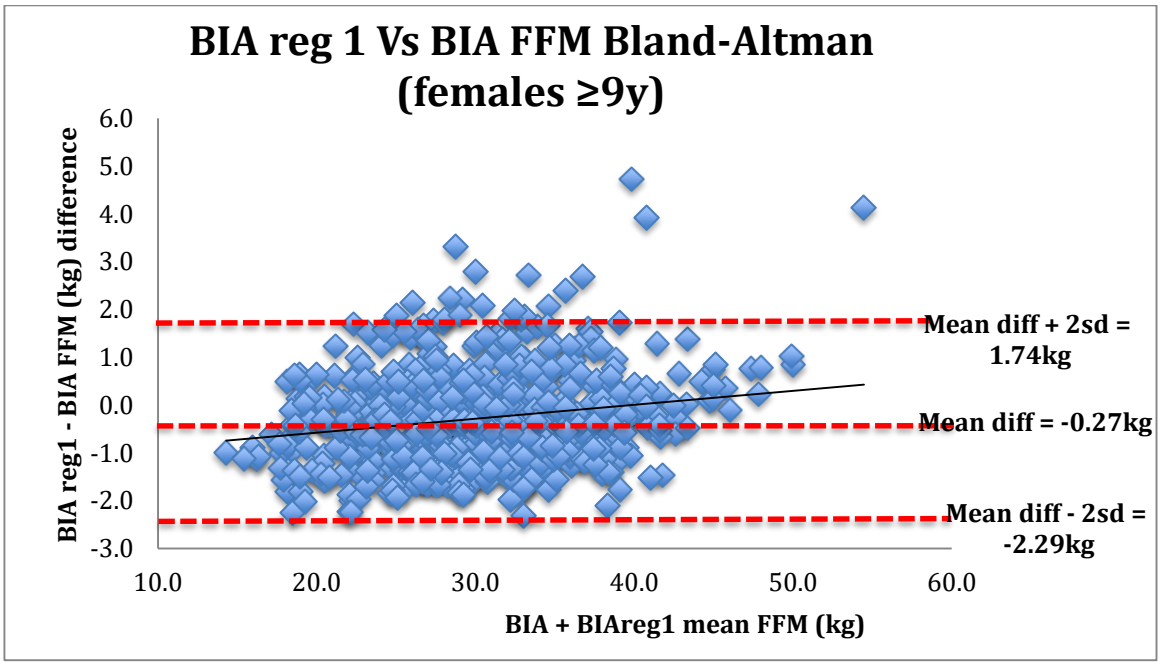
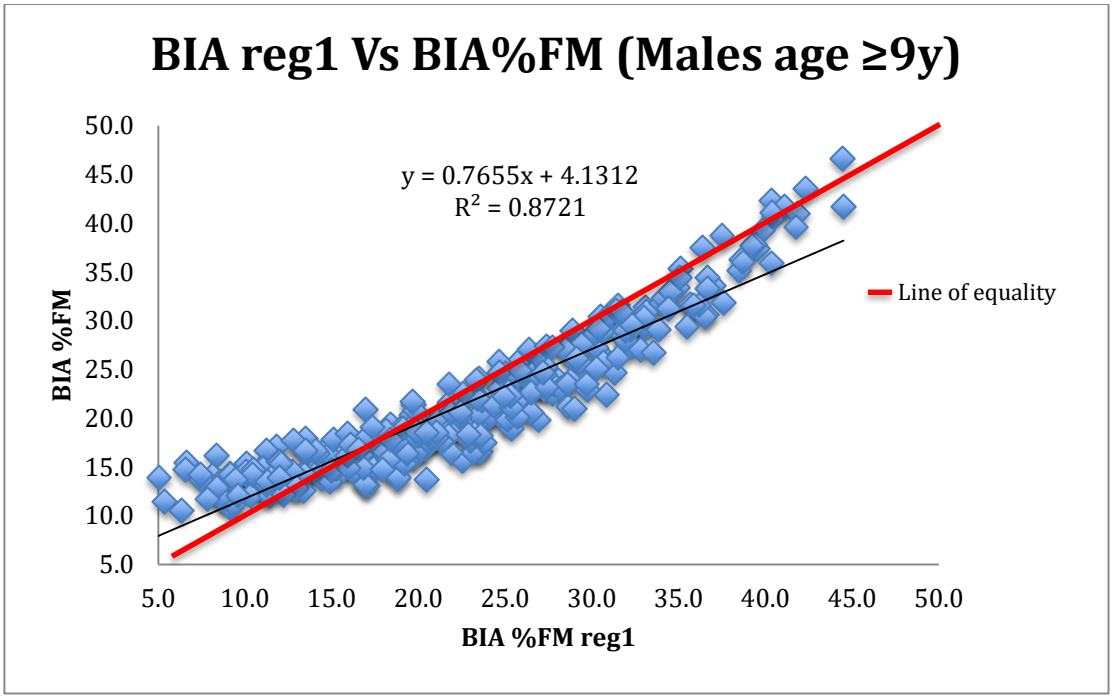


Figure 4.28 Scatterplots showing correlations (upper graph), and Bland-Altman limits of agreement showing mean differences (lower graph) by gender, between BIA FFM before correction and BIA FFMreg1 (BIA FFMreg1 = new BIA FFM following regression as described in section 4.6.3.4). BIA FFMreg1 = new BIA FFM following regression (see section 4.5.3.5); FFM = fat-free mass; DXA = dual-energy x-ray absorptiometry; BIA = bioelectrical impedance analysis.


BIA reg1 Vs BIA%FM (Males age ≥9y)
y = 0.7655x + 4.1312
R² = 0.8721
Line of equality
BIA %FM
BIA %FM reg1
50.0
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
5.0
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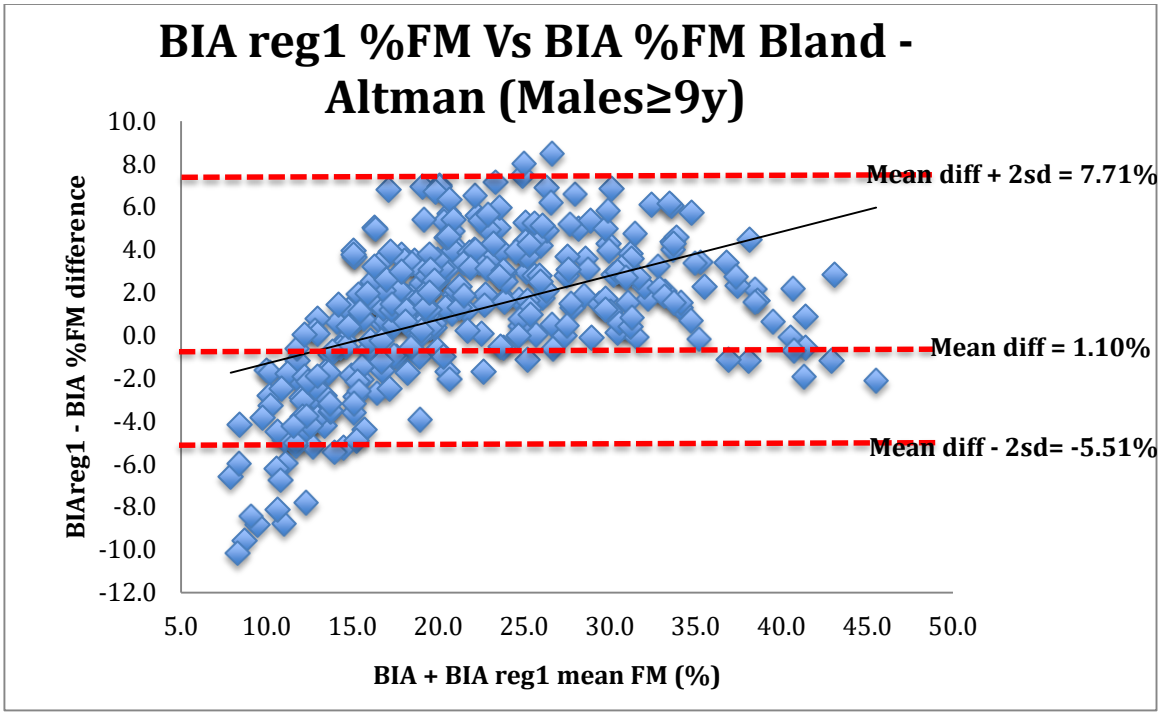
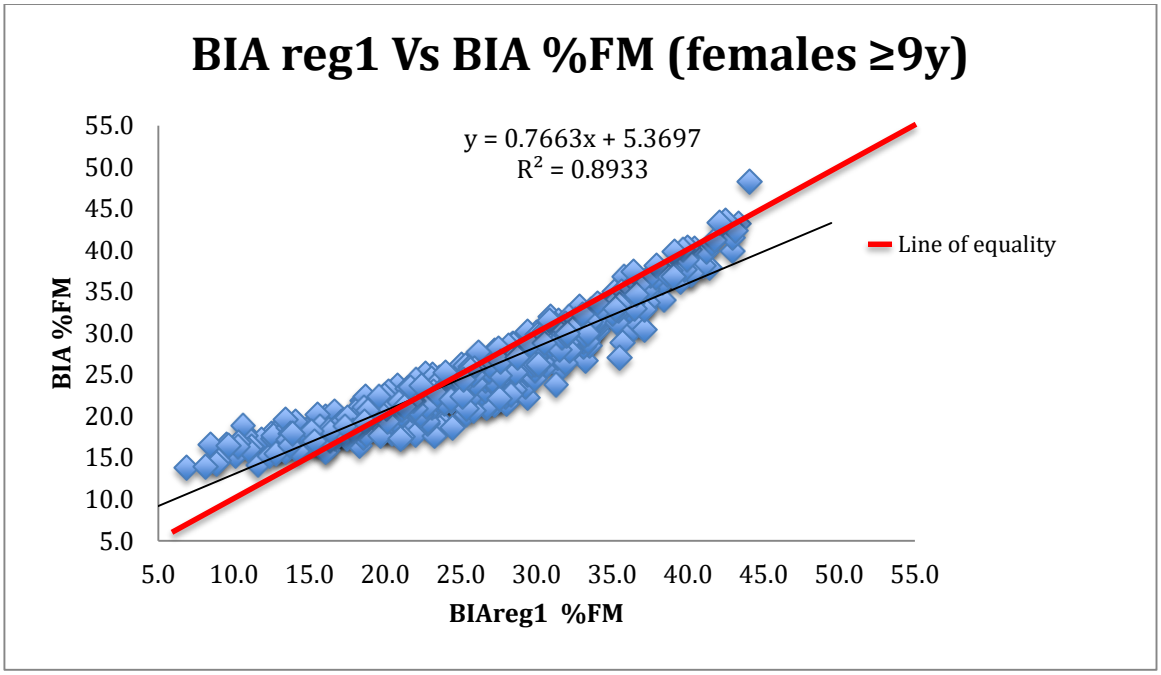
25.0
30.0
35.0
40.0
45.0
50.0


BIA reg1 %FM Vs BIA %FM Bland - Altman (Males≥9y)
Mean diff + 2sd = 7.71%
Mean diff = 1.10%
Mean diff - 2sd= -5.51%
BIAreg1 - BIA %FM difference
BIA + BIA reg1 mean FM (%)
10.0
8.0
6.0
4.0
2.0
0.0
-2.0
-4.0
-6.0
-8.0
-10.0
-12.0
5.0
10.0
15.0
20.0
25.0
30.0
35.0
40.0
45.0
50.0



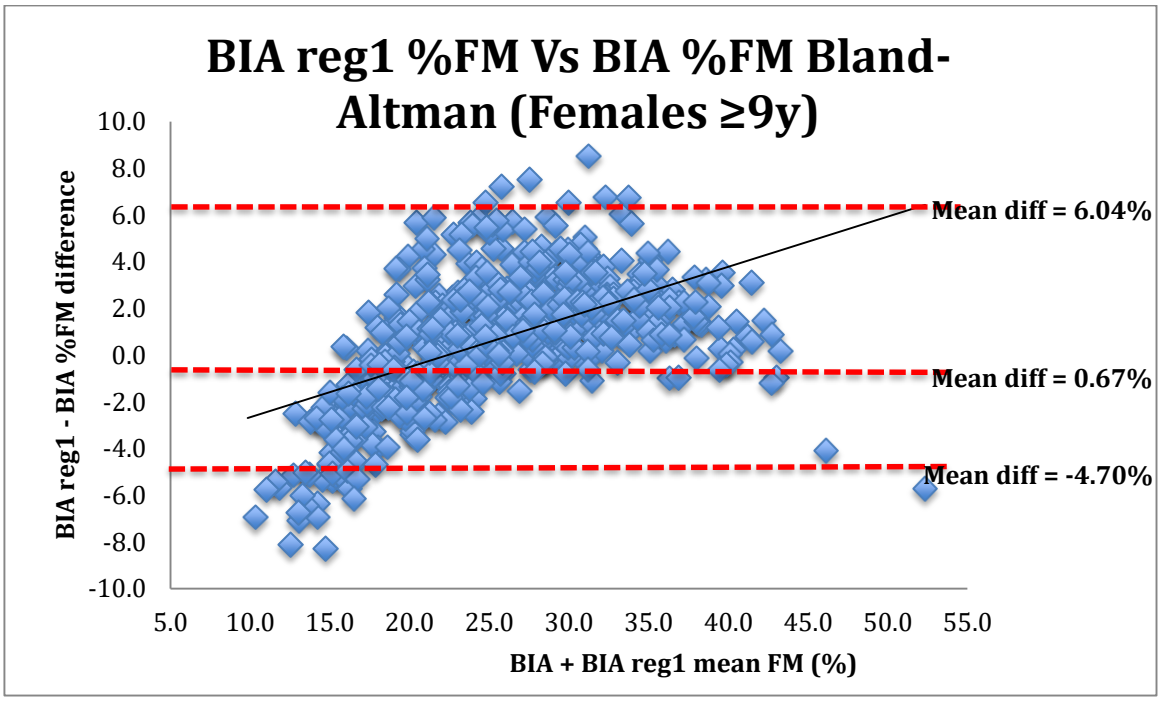


Figure 4.29 Scatterplots showing correlations (upper graph), and Bland-Altman limits of agreement showing mean differences (lower graph) by gender, between BIA %FM before correction and BIA %FMreg1. BIA FMreg1 = new BIA FM following regression (see section 4.5.3.5); FM = fat-free mass; DXA = dual-energy x-ray absorptiometry; BIA = bioelectrical impedance analysis.

4.5.3.8 Evaluation of FFM prediction equations from previous studies

Two previous studies that had developed SA prediction equations (Haroun et al., 2010; Sluyter et al., 2010) for FFM were also assessed against the study group using

the DXA_{Siri} FFM2 values, within the appropriate adolescent age ranges. BIA FFM_{reg1} was also compared to the DXA_{Siri} FFM2 values, within the specified age ranges for both studies, to assess how restricting the age range, would impact the new regression equation. The FFM prediction equations developed by Sluyter et al. (2010), using DXA as the reference method were:

i) $(0.607 \times H^2/Z) + (1.542 \times A) + (0.220 \times H) + (0.096 \times W) - 47.547$ for males

ii) $(0.531 \times H^2/Z) + (0.182 \times H) + (0.096 \times W) - 15.782$ for females

H = height (cm); Z = impedance (Ω); A = Age (years); W = weight (kg) the equations also included a value for ethnicity, which for Asians was zero, and thus ignored.

Haroun et al. (2010) used D_2O as the reference method, and developed the following regression equations for predicting total body water (TBW):

i) $-1.822 + (0.665 \times H^2/Z) + (1.288 \times \text{Asian})$ for males

ii) $0.125 + (0.647 \times H^2/Z) + (1.465 \times \text{Asian})$ for females

The equations also included a dummy variable for black ethnicity, which was ignored, as it was assigned a zero value for Asians.

Hydration of FFM was determined using the following equations based on a previous unpublished study (reported by Haroun et al., 2010):

i) $78.176 - (0.237 \times \text{age})$ for males

ii) $79.797 - (0.385 \times \text{age})$ for females

FFM was calculated as TBW/hydration of LM.

Correlations and Bland-Altman mean differences for both these equations, together with the outcomes for BIA_{reg1} from this study, are presented in Table 4.8. Application of both equations to correct BIA FFM, developed by Sluyter et al. (2010) and Haroun et al. (2010), resulted in significant differences between DXA_{Siri} FFM2 and BIA FFM,

for both sexes, with both equations significantly underestimating FFM, compared to DXA_{Siri}.

The equation developed by Haroun et al. (2010), underestimated FFM by the greatest amount, with much larger limits of agreement. Alternately, the equation developed in this study, even within the narrower age ranges, revealed no significant differences. In the 12-19y age range, BIA_{reg1}, underestimated FFM by a mean of 0.92kg (P = 0.229) in males and overestimated FFM by 0.78kg in the female sample (P = 0.147). In the 11-15y age range, BIA_{reg1}, underestimated FFM by a mean of 0.72kg (P = 0.319) in males, and overestimated FFM by 0.39kg (P = 0.518) in females.

Table 4.8 Comparisons (mean ±sd) between DXASiri FFM2 and BIA FFM regression equations from two validation studies with SA adolescent groups showing mean differences and Bland-Altman limits of agreement by age-range

Variables	Boys					Girls				
	N	**Age range (y)**	**Mean (kg) ± sd**	**Mean difference (DXA_{Siri} – BIA method) ± sd, (r) (kg)**	**Limits of agreement (mean ± 2sd; kg)**	**n**	**Age range (y)**	**Mean (kg) ± sd**	**Mean difference (DXA_{Siri} – BIA method) ± sd, (r) (kg)**	**Limits of agreement (mean ± 2sd; kg)**
DXA_{Siri} FFM2 (study group)	13	12-19	42.90 ± 14.27	-	-	15	12-19	35.18 ± 5.46	-	-
BIA FFM[1] (study group)	13	12-19	39.36 ± 14.90	3.54** ± 2.36, 0.98	-1.18 to 8.26	15	12-19	34.04 ± 4.55	1.14* ± 2.42, 0.95	-3.71 to 5.98
BIA FFM_{reg1} (study group)	13	12-19	41.98 ± 13.45	0.92 ± 2.62, 0.98	-4.32 to 6.16	15	12-19	35.96 ± 5.26	-0.78 ± 1.98, 0.93	-4.09 to 3.30
DXA_{Siri} FFM2 (study group)	13	11-15	34.69 ± 12.29	-	-	14	10-16	32.24 ± 6.83	-	-
BIA FFM[2] (study group)	13	11-15	27.79 ± 8.91	6.90** ± 4.12, 0.97	-1.35 to 15.16	14	10-16	28.17 ± 5.16	4.07** ± 2.72, 0.95	-1.36 to 9.51
BIA FFM_{reg1} (study group)	13	11-15	33.97 ± 11.11	0.72 ± 2.51, 0.98	-4.30 to 5.75	14	10-16	32.62 ± 7.12	-0.39 ± 2.09, 0.96	-4.56 to 3.78

DXA = Dual-energy x-ray absorptiometry; BIA = Bioelectrical impedance analysis; FFM = fat-free mass; BIA FFM[1] = Sluyter et al. (2010) regression equation to predict BIA FFM; BIA FFM[2] = Haroun et al. (2010) regression equation to predict BIA FFM

** P< 0.001 Sig. (2-tailed); * P< 0.05 Sig. (2-tailed); BIA FFM_{reg1} = BIA corrected following regression (see section 4.5.3.5)

4.6 Comparisons between BIA & DXA SMMa

SMMa data for DXA and BIA were compared for both absolute (kg) and relative (%; SMMa (kg)/body weight (kg) x 100). Whilst correlations between the two methods for both absolute and relative SMMa were strong (r =0.97 and 0.72 respectively), the mean differences were significant but small (mean differences = 1.28kg & 1.52%; P<0.001 & P = 0.001 respectively) with BIA underestimating SMMa (see Table 4.9). However, due to the differences between DXA Wt and BIA scale Wt (see section 4.6.2), making adjustments to correct DXA for limb Wt was not possible. Furthermore, adjustment of the markers to selectively incorporate all four limbs using DXA software was difficult and required subjective judgement, particularly where participants were lying with legs together or arms in contact with the body. Additionally, some participants did not fully fit within the scanning area. Therefore, correction for BIA SMMa was not conducted.

Table 4.9 Descriptives showing absolute (kg) and relative (%) BIA and DXA SMMa (mean ±sd), with mean differences for SA BIA validation study group

Variables (n=51)	**Mean ± sd**	**Mean difference (DXA – BIA)**
DXA SMMa (kg)	14.13 ± 6.55	1.28**
BIA SMMa (kg)	12.85 ± 6.12	-
DXA SMMa (%)	30.66 ± 4.45	1.52**
BIA SMMa (%)	29.14 ± 3.71	-

BIA = Bioelectrical impedance analysis; DXA= Dual-energy x-ray absorptiometry; SA = South Asian; SMMa = Appendicular skeletal muscle mass; ** Significant differences at P< 0.001 level Sig. (2-tailed)

4.6.1 Discussion

This study provides an improved BIA (for the BC418-MA model) prediction equation validated against DXA (Norland Pencil-beam scanner) for estimating absolute FFM (kg), from which %FM can be determined for the SA child (aged 9y – 18y) and adolescent population. Whilst the aim of the study was to develop a prediction equation for all SA children and adolescents aged from 5-18y, this proved to be difficult, as when the new equation was applied to the SA external group, spurious outcomes for children <9y were observed. This was attributed to the significant ($P< 0.05$) differences in age between the validation study group (mean age = 12.84y & 13.5y) and the SA external group (mean age = 9.12y & 10.00y) for boys and girls respectively. In a recent 4-C study on BIA, it was concluded that a single equation to determine FFM across the whole child and adolescent age range (4-24y) would not be valid as at different pubertal stages the slope and intercept of the regression between Ht^2/Z and FFM changes (Montagnese et al., 2013). Due to the opportunistic nature of recruitment for the validation study, age selection, particularly at younger ages was difficult.

The analysis conducted on %FM between the three methods confirmed that in general BIA significantly underestimated body fat when compared to DXA (Figures 4.7 to 4.11) and ADP (Figure 4.18). The new prediction equation developed in this study reduced the mean %FM differences (non-significant) between BIA and DXA in both boys and girls, with BIA_{reg1} now overestimating %FM in boys by 1.07%, and underestimating %FM by 0.73% in girls (Table 4.6). Whilst there was a strong positive correlation between DXA and ADP with a small mean difference, the 95% limits of agreement were very large (Figure 4.19), with a significant difference between the two measurement methods, with ADP underestimating %FM compared to DXA. Other studies have reported similar finding when comparing these two technologies, and advised against using these methods interchangeably (Hames et al., 2014).The large limits of agreement observed between the ADP and DXA, may also have been due to the fact that a number of the participants were not willing to wear skin-tight swimsuits due to cultural and religious sensitivities, and whilst long-sleeved swim tops and leggings were provided they were not entirely skin-tight; this may have contributed to increased body volume, leading to errors in

body composition estimation. Thus, further analyses were only conducted with BIA and DXA.

This new prediction equation was compared to equations developed in two other BIA validation studies (Haroun et al., 2010; Sluyter et al., 2010) for SA child and adolescent populations with more restricted age ranges (see section 4.5.3.8), against DXA_{Siri} FFM_2. Both equations resulted in BIA significantly underestimating FFM (kg) compared to DXA (Table 4.8), with the greatest difference observed between the equation developed by Haroun et al. (2010). However, the prediction equation developed in this study resulted in no significant differences between BIA and DXA_{Siri} FFM_2 within the same age ranges specified in the respective studies. The much larger differences between the equation developed by Haroun et al. (2010), may have been due to the fact that that validation study used a leg-leg BIA model (Tanita TBF-300) validated against deuterium (D_2O); whilst the study conducted by Sluyter et al. (2009) used the same BIA model (Tanita BC-418) as in this study, and also used a DXA pencil-beam scanner (GE Lunar) as the reference method. Variations in FFM estimation between DXA models have been reported in review studies (Helba & Binkovitz, 2009; Plank, 2005), which may also be a contributory factor in the differences between this study and the study conducted by Sluyter et al. (2010).

Studies comparing %FM with tetrapolar BIA devices against reference methods including deuterium (D_2O) and DXA have not found a significant mean bias between the two methods (Bosy-Westphal et al., 2008; Pietrobelli et al., 2004). However, one study involved a Caucasian population (Bosy-Westphal et al., 2008) and the other study which also used the Tanita BC418 model with DXA as the reference method, did not involve an ethnic-specific group (Pietrobelli et al., 2004). In this study, the large differences between DXA and BIA are likely attributed to the SA ethnicity of the sample, and supports the need to validate BIA models and develop ethnic-specific prediction equations, as evidenced by other research, in which ethnic differences in hydration levels of FFM is recognised (Frisard et al., 2005; Jebb et al., 2000; Sun et al., 2003; Rush et al., 2006; Williams et al., 2007).

In this study, whilst BIA prior to correction underestimated %FM overall, there was a tendency to overestimate %FM at the leaner end of the scale and underestimate %FM at high adiposity levels for both sexes. This tendency was reduced following correction (see Figure 4.30), as was observed when the original BIA equation was compared to the new equation. Underestimation of FM in leaner subjects and overestimation at high body fat levels, has also been observed in other studies (Pietrobelli et al., 2004; Sluyter et al., 2010), in which the same BIA (BC-418) instrument was used. Other studies have reported greater bias in %FM with bipolar BIA instruments than with tetrapolar instruments, with a similar overestimation of %FM in lean subjects and an increasing underestimation of %FM as adiposity increased, in all bipolar devices (Bosy-Westphal et al., 2008; Mitsui et al., 2006).

For both the validation and external group, mean BMI SDS was lower than the UK90 data, whilst %FM and WC SDS values were higher (see Table 4.1). Furthermore, considering that the SDS values for %FM were derived from the original uncorrected BIA data, which was found to underestimate %FM, highlights the inadequacy of BMI as a reliable indicator of adiposity and supports the considerable body of evidence that other more direct measures of adiposity are required in addition to BMI, particularly in the high risk SA population (Haroun et al., 2010; Sluyter et al., 2010).

Whilst BIA was found to significantly underestimate SMMa for both absolute (kg) and relative (%) values (section 4.6), due to differences in DXA Wt and scale Wt (see section 4.5.2) it was not possible to correct DXA limb Wt. Additionally, it is important to note that compared to DXA, BIA was found to consistently underestimate %FM as stated above, and thus by extrapolation, any underestimation of SMMa would be likely due to DXA sum of parts and scale Wt differences, where DXA Wt was greater than scale Wt. Furthermore, the differences between DXA and BIA whilst statistically significant were in reality very small with a relative difference of only 1.52%. It was therefore considered acceptable to use BIA for estimating SMMa without additional correction.

4.6.2 Limitations

Due to the opportunistic nature of recruiting for the validation study, the equation produced was not applicable to children <9y of age. This study would need to be repeated by targeting children aged 5-9y and developing another prediction equation for this age range. No single BIA equation is likely to be valid throughout childhood and adolescence due to changes in body proportions (Montagnese et al., 2013), and furthermore, the hydration of FFM is not constant during this period (Lohman, 1989; Montagnese et al., 2013; Wells et al., 2010). Whilst the mean differences for absolute FFM and %FM between DXA_{Siri} and BIA_{reg1} for both sexes were not significant, the limits of agreement were fairly large, as has been reported in other studies (Wells et al., 1999), which indicates that BIA can give accurate results for epidemiological purposes, but body composition estimations at an individual level should be treated with caution, and preferably in conjunction with other measures such as BMI and WC. Assessment of body composition variations within the SA ethnic group could not be conducted due to sample size limitations.

Data from ADP (BodPod) measurement could not be used due to the very large limits of agreement between DXA and BodPod, with possible procedural errors, attributed to swim attire not being sufficiently skin-tight. Whilst DXA is regarded as a valid and reliable reference method for body composition assessment, variations between models have been reported in review studies (Helba & Binkovitz, 2009; Plank, 2005). A standardised approach to using DXA for body composition assessment has been called for in expert reviews (Lohman & Chen, 2005; Plank, 2005), although variation in %FM estimates between pencil-beam scanners is estimated to range between 1-3% (Lohman & Chen, 2005). In this study, differences in scale Wt and DXA sum of parts (see section 4.6.2) had to be corrected for using relative (%) FM values; a factor observed in other studies (Friedl et al., 1994; Nelson et al., 1996; Korth et al., 2007). Clearly, further research is required to investigate this issue.

Many experts recommend the 4-C model as the criterion method for *in-vivo* body composition assessment (Going, 2005; Wells & Fewtrell, 2006). However, this method requires use of multiple methodologies that are prohibitive in many

research settings (Lee & Gallagher, 2008; Wells et al., 2012). A recent study on body composition in children and adolescents, revealed very high correlations in FM and FFM SDS values between the 4-C model and DXA (Wells et al., 2012).

4.6.3 Conclusion

This study provides a robust ethnic-specific BIA prediction equation suitable for SA children and adolescents (aged ≥9y) that has been validated against DXA using the BIA tetrapolar device (Tanita BC-418). However, the equation needs to be tested in an independent group to confirm its accuracy and functionality, before it can be applied to this BIA model. The results of this study confirm that the proprietary in-built equation significantly underestimates %FM for the SA population. Due to fairly large limits of agreement between DXA and BIA, body composition assessment with BIA, at an individual level, should be treated with caution. BIA in conjunction with WC offers a more objective measure of adiposity than BMI, particularly for the SA ethnic group, where BMI has been shown to be inadequate. The importance of conducting large-scale body composition assessment, particularly for the SA community, is well documented, and so it suits our needs to have a portable and valid BIA system that can be taken into the field.

CHAPTER 5: Ethnic Differences in Body Composition Among South Asian & White European Children & Adolescents

5.1 Introduction

There is much evidence for ethnic variations in body composition (BC) between South Asian (SA) and white European (WE) children (Donin et al., 2010; Ehtisham, et al., 2005; Haroun et al., 2010; Mehta et al., 2002; Misra et al., 2003; Rush, 2004; Shaw et al., 2007; Stanfield et al., 2012; Taylor et al., 2005; Whincup et al., 2007; Whincup et al., 2010) and adult (Dulloo et al., 2010; Lear et al., 2007; Misra et al., 2007; Whincup et al., 2010) populations both in the UK and worldwide (Liu et al., 2009; Whincup et al., 2010); with SAs tending to have higher % body fat and visceral fat and less lean mass (LM) at equivalent body mass index (BMI) values (Lear et al., 2009; Saxena et al., 2004; Viner et al., 2009). Whilst BMI (BMI centiles for children and adolescents, and BMI cut-offs for adults), is a proxy measure for determining adiposity (see Chapter 2 section 2.3), and is widely regarded as an unreliable indicator of health for the SA population (Dulloo et al., 2010; Saxena et al., 2004; Viner et al., 2009), it is the most established classification method for overweight and obesity, used both in the UK (NHS Information Centre, 2011; NICE, 2006) and worldwide (WHO, 2000).

The differences in BC underpinned by a genetic predisposition to the metabolic syndrome, are considered to be the underlying causes for the higher risk and prevalence of type 2 diabetes mellitus (T2DM) and cardiovascular disease (CVD) within this population group, when environmental factors are accounted for (Bhardwaj et al., 2008; Chaturvedi, 2003; Misra et al., 2007; Saxena et al., 2004; Whincup et al., 2010), with SAs compared to WEs, described as having a 'thin-fat' phenotype evident from birth (Gholap et al., 2011; Yajnik et al., 2002; Yajnik et al., 2004). The overall body size of SAs compared to WEs is also smaller from birth (Krishnaveni et al., 2005) and in adulthood (Eveleth & Tanner, 1990; Rush et al., 2009) with short-stature in adulthood associated with an increased risk of obesity (Guerrero-Igea et al., 2001; Gunnell et al., 2003; Lawlor et al., 2002). The 'thrifty phenotype' hypothesis (Hales & Barker, 2001; see Chapter 2, section 2.6) has been cited as a possible cause for a shorter stature and lower body weight (Wt) at birth

(Bogin & Valera-Silva, 2010), with rapid infant Wt gain regarded as an increased risk for developing T2DM in adulthood (Hales & Barker, 2001).

To account for the increased health risks to SAs for T2DM and CVD, lower BMI adult cut-offs have been advised as 'action' or' 'trigger' points (Misra et al., 2009; WHO, 2004; see also Chapter 2, section 2.3.1). Additionally, due to the increased health risks associated with abdominal adiposity, sex-specific waist circumference (WC) cut-offs are also used as an additional measure for adults (IDF, 2006; WHO, 2011), with India adopting lower WC cut-offs for its indigenous population (Misra et al., 2009), and the IDF (2006) also advising a lower WC cut-off for SA men. However, in the UK, NICE (2006, 2013), has not adopted lower BMI or WC cut-offs for SAs, but advised the use of 'caution and clinical judgement' by healthcare professionals. For children, both internationally (Cole et al., 2007; de Onis et al., 2007) and in the UK (Cole et al., 1995), only BMI percentile charts are used for defining overweight and obesity, although the IDF (Zimmet et al., 2007) advises the use of WC percentile cut-offs for determining abdominal obesity and diagnosing metabolic syndrome (see Chapter 2, section 2.3.2). NICE (2013), also recommended (as with SA adults), in the absence of ethnic-specific growth reference charts, the use of 'caution' when interpreting BMI SDS for SA and other minority ethnic groups, and advised that the use of WC may be used as an additional measure if necessary.

Adiposity alone is not the main contributor to T2DM or insulin resistance, particularly among SA children within the normal Wt and BMI ranges (Lear et al., 2009; Whincup et al., 2002; Whincup et al., 2010). The risk of mortality increases with increasing percentage fat mass (FM) and decreasing fat-free mass (FFM) relative to height (Ht; FFMI; FFM (kg)/Ht (m^2)) and therefore such measures are recommended in addition to BMI, in clinical settings (Bosy-Westphal et al., 2009; Cole et al., 2000; Dulloo et al., 2010; McCarthy et al., 2003; Sun et al., 2003). In a recent school-based Child Heart and Health Study (CHASE; Whincup et al., 2010), involving 4,796 healthy children aged 9-10y, from 200 UK schools, which included a large number of SA children (n= 1,306), compared to WE children the SA cohort had similar Ht, lower Wt, mean ponderal index (Wt kg/Ht m^3), and WC, but a higher FM index (FMI; FM (kg) /Ht (m^2)) and mean sum of four skinfolds. However,

adjustment for adiposity had little effect on the blood markers related to the risk of T2DM. In a study conducted by Lear et al. (2009) comparing BC differences between third generation Canadian male and female immigrants of SA, Aboriginal, Chinese, and European origin (mean age 44-50.7y), after adjustment for possible confounding variables (age, Ht, frame size, sociodemographic and lifestyle factors), SAs had significantly less LM at any given FM value, with a higher FM to LM (F: LM) ratio. Other studies have reported similar findings, with SAs having less LM and equivalent %FM as other ethnic groups at lower BMI values (Rush et al., 2009).

Further research has been recommended into the differences in body composition between SA and WE children to evaluate the potential risks associated with these differences (Viner et al., 2010). In a report on research priorities related to diabetes for British SAs published by Diabetes UK and the South Asian Health Foundation (Gholap et al., 2009) highlighting gaps in the research; one of the priorities listed for SA children and adolescents was "determining the most effective screening method for Type 2 diabetes in children and how this is affected by ethnicity" (p.55).

FFM or skeletal muscle mass (SMM), in particular appendicular SMM (SMM_a) is regarded as an important indicator of health, being the primary site for insulin mediated glucose disposal (Dulloo et al., 2010; McArdle et al., 2007; McCarthy et al., 2014; Miller et al., 1984), as well as active in fat metabolism (Seidell et al., 2001). Assessment of SMM_a, had until recently been restricted to complex laboratory assessment methods such as magnetic resonance imaging (MRI) and dual-energy x-ray absorptiometry (DXA), however tetrapolar bioelectrical impedance analysis (BIA) models such as the Tanita BC-418MA offer a quick, simple, safe and cost-effective means of taking such measures in addition to body fat estimation (McCarthy et al., 2013; Pietrobelli et al., 2004; see chapter 2 Literature Review section 2.3.1 for further details).

SMMa relative to total body FM (i.e. SMMa (kg)/FM (kg), defined as the muscle-to-fat ratio (MFR; McCarthy et al., 2014), has also been identified as a useful measure, when making between-group comparisons. The theory behind use of the MFR is based on the understanding that muscle tissue is much more active in glucose

metabolism with greater sensitivity to insulin compared to fat tissue which inhibits insulin action and sensitivity by the release of free-fatty acids, thus in addition to high levels of adiposity, a low MFR would indicate increased risk of metabolic syndrome (Kim et al., 2011). Use of a fat and fat free mass index (FMI and FFMI respectively) has also been recommended as a more useful comparison of BC between individuals of different heights than BMI or %FM, as it is considered less likely to misclassify highly muscular individuals as overweight or obese, and would also help to identify individuals and populations that may be at risk of T2DM due to a high FMI or low FFMI that fall within the normal BMI range (Dulloo et al., 2010).

Within the UK SA population, the prevalence of T2DM is reported to be higher among Pakistanis and Bangladeshis than Indians (Bhopal et al., 1999; Erens et al 2001; Mindell & Sproston, 2006; Wild et al., 2007), with some studies reporting Bangladeshi children having a more adverse health profile than Pakistani and Indian children in relation to T2DM (Whincup et al., 2010), which is consistent with reports of the very high prevalence of T2DM among Bangladeshi adults (Bhopal et al., 1999; Erens et al 2001; Mindell & Sproston, 2006). Therefore, where possible, within group comparisons of the measured variables of associated risk factors are recommended, to gain an increased understanding of these differences in health outcome. Although there is evidence that these differences may be attributed to lifestyle and socioeconomic factors (Bhopal et al., 1999; Magnhild et al., 2008; Murasko, 2009; Owen et al., 2009; Saxena et al., 2004).

BIA provides a relatively accurate, safe, simple, and non-invasive method of estimating BC, including relative (%) and absolute (kg) FM and FFM. The Tanita (BC-418, Tokyo) model, also provides whole body and segmental estimates for FM, FFM, and segmental predicted muscle mass (PMM) for all four limbs, as well as the trunk and head (Pietrobelli et al., 2004; McCarthy et al., 2006), and has been validated against reference methods including DXA (Pietrobelli et al., 2004) and deuterium oxide (D_2O; Bosy-Westhphal et al., 2008) in both child and adolescent populations. However, neither of these studies was specific to the SA child and adolescent ethnic group, and due to ethnic variations in hydration of FFM (Heyward & Wagner, 2004; Kyle et al., 2004; Lohman, 1989), ethnic-specific prediction equations are

recommended (Chumlea & Sun, 2005; Dehghan & Merchant, 2008; Haroun et al., 2010; Kyle et al., 2004; Rush et al., 2006; Ward, 2012; Williams et al., 2007).

To address the lack of a valid BIA prediction equation for %FM for the SA child and adolescent population, a BIA validation study using DXA and air displacement plethysmography (ADP) as reference methods was conducted to develop ethnic-specific prediction equations for this population group (see Chapter 4). Consistent with other studies on SA populations (Haroun at al., 2010; Sluyter et al., 2010), using the in-built proprietary equations, BIA compared to DXA, was found to significantly ($P < 0.05$) underestimate %FM by a mean of 2.23% and 2.93% in boys and girls respectively. The successful development of a prediction equation (Chapter 4) reduced this difference to 1.07% and 0.73% in boys and girls respectively. However, as the cohort for the validation study had a very limited number of children < age 9y, the equation developed was not applicable to children below this age.

In this study, comparisons between DXA and BIA for SMMa both absolute (kg) and relative (%) were also made (see Chapter 4 section 4.7), which revealed BIA was significantly underestimating SMMa ($P< 0.001$). However, as DXA Wt was significantly greater than scale Wt (see Chapter 4 section 4.6.2), and DXA limb Wt could not be corrected for, this difference was not considered to be a true indication of SMMa differences, particularly when BIA was found to be significantly underestimating %FM. Whilst the differences were statistically significant, due to the very small relative differences in %SMMa (mean difference DXA – BIA = 1.52%) between BIA and DXA, it was considered acceptable to use BIA SMMa data without further correction. There were also difficulties using the DXA software in subjectively marking out the four limbs, particularly where the participants' arms and legs were in contact with the body. Furthermore, some very large subjects did not fit within the scanned area.

Due to the known limitations of BMI (see chapter 2 section 2.3), McCarthy et al. (2006), developed sex-specific body fat reference curves for UK children of WE origin, based on a cohort of 1,985 children aged 5-18y. These percentile curves had defined cut-offs for 'underfat', 'normal', 'overfat' and 'obese', set at the 2^{nd}, 50^{th}, 85^{th},

and 95th percentiles respectively, to match the IOTF BMI cut-offs (Cole et al., 2000). In that study, BC was assessed using segmental BIA (Tanita BC-418MA). These charts have been published by the Child Growth Foundation (CGF), for use in clinical practice, alongside BMI. The authors recognised the limitation of having defined cut-offs for %body fat in children and adolescents similar to BMI cut-offs that are not underpinned by evidence related to health risk, as many of the obesity-related health risks become apparent in adulthood. However, based on evidence that obesity levels among children are rising and have a tendency to track into adulthood, and T2DM is no longer an adult disease, particularly among SAs, there is a clear need to have specific measures for BC that go beyond BMI.

Following the publication of body-fat percentile curves for WE children and adolescents (McCarthy et al., 2006), McCarthy et al. (2013) also published sex-specific SMM_a reference curves, using the SMMa data from the same children, which involved summing the SMM (kg) predicted output for all four limbs (i.e. SMM_a) taken from BIA (Tanita BC-418MA) and converting it to relative (%)SMM_a ($\%SMMa = \frac{SMMa(kg)}{body\ weight(kg)} \times 100$.

There are currently no body fat or SMM_a percentile charts available for the SA child and adolescent population. Based on the evidence of higher levels of % body fat and lower LM, between SAs and WEs for a given BMI (Gray et al., 2011; Liu et al., 2011; Luke, 2009; Mehta, 2002; Misra et al., 2009; Rush, 2004; Taylor et al., 2005; Whincup et al., 2007; WHO, 2004), there is a clear need to have ethnic specific body composition data, with body fat and SMM reference curves for SAs with appropriate cut-offs to identify those children that may be at increased risk of developing obesity-related diseases (Misra et al., 2005; Saxena et al., 2004). The development of body fat and SMMa percentile curves specific to SA children and adolescents, similar to those developed by McCarthy et al. (2006 & 2013 respectively) for WEs will fill an important gap in the research.

5.1.1 Study Aims

The aims of this study were to:

i) Develop sex-specific %FM reference curves for the SA child and adolescent (9-18y) population in the UK, by applying the new prediction equations developed in Chapter 4.

ii) Develop a range of sex-specific SMM_a reference curves for the SA child and adolescent (5-18y) population in the UK for use in epidemiological and clinical settings as an additional measure to BMI and WC.

iii) Assess within-group variations for %FM and $\%SMM_a$, between Indian, Pakistani, and Bangladeshi children and adolescents.

iv) Compare the new reference curves for SA children and adolescents with the WE %FM (McCarthy et al., 2006) and various SMM_a (McCarthy et al., 2013) reference curves, with main comparisons with available WE low income datasets.

v) Evaluate the differences between SA and WE children and adolescents in %FM and $\%SMM_a$ and compare these differences to the BMI UK90 reference standards.

5.2 Participants and Methods

5.2.1 Participants

Data from 1,459 (n = 584 boys and 875 girls) SA school-aged children from London boroughs measured on the same Tanita (BC418-MA) analyser from a previous study conducted at this university (from 2005 to 2007) was used in this study, together with additional data (n =121; 39 girls and 82 boys) from a further field-based study that was conducted to add to the Pakistani data set (see Chapter 4 section 4.6.1).

A large majority of the SA children were from more deprived areas of inner city London such as Tower Hamlets, whilst the WE_1 dataset used in both studies in which

%FM centiles and SMMa centiles were produced (McCarthy et al., 2006 & 2013 respectively) involved children that were intentionally selected from more affluent backgrounds to minimise the effect on the outcome of rising levels of obesity in order to make more valid comparisons with the UK90 reference population. Thus to ensure that comparisons between the WE_1 and SA sample were not overly influenced by socioeconomic status, additional comparisons for %FFM and SMMa were made between only the 'low income' WE (WE_2) data set. For sex-specific within-group comparisons, SA boys and girls were divided into Indian, Pakistani, Bangladeshi, and other Asian (i.e. of mixed SA heritage) sub-groups based on self-report. Within-group comparisons were restricted to the Indian, Pakistani, and Bangladeshi sub-groups as the aims of this study were limited to these sub-groups, which are commonly referred to in other research.

5.2.2 Methods

Anthropometric measures of Ht, Wt, BMI, and SMMa were taken from the original Tanita BIA (BC418-MA) printouts from the existing dataset of SA children, and WC measures were taken from written records. Two field workers on school premises took these measurements (McCarthy et al., 2006). The additional data taken from the field-based study (Chapter 4) was added to this dataset, and followed the same measurement protocol described by McCarthy et al. (2006) in order to minimise measurement error. Methods used for all anthropometric and BIA measures are also described in chapter 3 (General Methods). BC measures included absolute (kg) and relative (%) whole body FM and FFM taken from the Tanita printouts. Both these measures were corrected for, using the new regression equation developed in the BIA validation study (chapter 4), which was limited to children aged ≥9y as explained in the introduction.

The LMS method (Cole & Green, 1992; see chapter 2 General Methods, section 3.4.2 for more information) was used to construct sex-specific centile curves for %FM and %FFM using the original BIA equation and new BIA regression equation for children aged ≥9y, to make comparisons between the results before and after regression, and also to compare the results with the WE_1 and WE_2 data. %FFM centile curves using the original BIA regression equation were also constructed for all children aged ≥5y to make comparisons with the WE_2 dataset across all ages. For SMM, only

appendicular SMM (i.e. SMMa) data from BIA was used to construct centile curves, as this accounts for the majority (~75%) of whole body SMM in adults (McCarthy et al., 2013), and has the potential of being modified through physical activity and exercise (McCarthy et al., 2013). SMMa measures included: absolute (kg); relative (%) SMMa (SMMa (kg)/body Wt (kg) x 100); SMMa/FFM% (SMMa (kg)/FFM (kg) x 100); SMMa Index (SMMaI = SMMa (kg)/Ht (m^2); and MFR (SMMa (kg)/ FM (kg)).

5.3 Statistical Analyses

Data for Ht, Wt, BMI, %FM, and WC were also converted to standard deviation scores (SDS), using LMS Growth software (see Chapter 3, section 3.4.2), to compare with UK90 reference data (Cole et al., 1995). As SDS references were not available for FFM or SMMa data, to enable comparisons to be made by age, the datasets were divided into the following age ranges: 5-7y; 8-10y; 11-13; 14-16y; and 17+ in accordance with the age ranges used by McCarthy et al. (2013).

Between-group (SA and WE) comparisons were conducted using Independent Samples T-tests, and within-group comparisons were conducted using ANOVA, with the level of significance set at the $P<0.05$ level unless otherwise stated. All analyses were conducted using the Statistical Package for Social Sciences (SPSS v.20 for Mac; IBM.com). Whilst absolute values within each age range were determined, reference was only made to SDS data where available (i.e. for Ht, Wt, BMI, %FM, and WC), as SDS adjusts for age more accurately than age ranges and also provide a comparison to the UK90 reference data. To make direct comparisons with WE_1 (McCarthy et al., 2006 & 2013) centiles, the same 9 centiles (i.e. 2, 9, 25, 50, 75, 85, 91, 95, 98) were selected for the BIA measured variables stated above and presented in corresponding centile charts, although for clarity the 75^{th} and 85^{th} centiles were excluded from the centile graphs.

5.4 Results

A total of 1,624 (691 boys aged 4.6-20.7y, and 933 girls aged 4.5-21.5y) SA and 1,141 (598 boys aged 4.5y and 543 girls aged 4.5 – 16.7y) WE_2 subjects comprised the total dataset above age 4.5y. However, due to very few numbers above age 14y in both the SA and WE_2 samples, the cut-off age for analysis was 14y. As the aim of this study

was to develop centile charts for children aged 5-18y, children above the age of 4.5y were included in the 5y age group and those below were excluded. The final dataset comprised 1581 (666 boys and 915 girls) SA and 1137 WE2 subjects (596 boys and 541 girls) aged 5 – 14y. Less data (SA n= 1366; WE2 n= 1135) was available for all SMMa measures due to missing data.

Table 5.1 provides sex-specific descriptive data, showing mean SDS values based on the UK90 reference data (Cole et al., 1995) for Ht, Wt, BMI, WC, and %FM for both the SA and WE2 (low-income) dataset as a whole. Compared to the UK90 reference population, SA boys and girls were on average shorter, lighter, with a lower BMI, but with a higher WC and %FM, whilst WE2 boys and girls had higher mean SDS values for all measures. T-test comparisons between the SA and WE2 cohort, revealed that both SA boys and girls were significantly (P<0.001) shorter, lighter, with a lower BMI and WC than their WE2 counterparts, with no significant differences in %FM.

Table 5.1 Mean ±sd SDS data for Ht, Wt, BMI, WC, & %FM for SA[a] and WE2[b] cohort with sex-specific comparisons between ethnic groups

Age (y)	n	Ht SDS	Wt SDS	BMI SDS	WC SDS	%FM SDS
Boys						
[a]9.01 ±2.60	666	-0.02** ±1.00	-0.08** ±1.37	-0.14** ±1.52	0.29** ±1.36	0.53 ±1.08
[b]8.84 ±2.43	594	0.29 ±1.00	0.44 ±1.25	0.37 ±1.35	0.74 ±1.17	0.49 ±1.07
Girls						
[a]10.00 ±2.56	912	-0.07** ±1.01	-0.15** ±1.31	-0.21** ±1.45	0.21** ±1.45	0.25 ±1.11
[b]8.81 ±2.39	541	0.15 ±1.02	0.22 ±1.20	0.18 ±1.27	0.64 ±1.19	0.25 ±1.07

Ht = height; Wt= weight; BMI = body mass index, WC= waist circumference; FM= fat mass; SDS = standard deviation score
**Significant difference at P<0.001 level
a= South Asians; b= white Europeans

Table 5.2 shows the sex-specific descriptive statistics for both the SA and WE2 cohort by age range, together with significant (at P< 0.05 and P< 0.001 levels) outcomes for between group t-test comparisons by sex and age-range. Absolute data for %FM is not presented, as this would be the reciprocal of % FFM. Compared to WE2 boys, SA boys in the age ranges 5-7y and 8-10y were significantly shorter and lighter, with a significantly lower BMI based on SDS data. In the 11-13y age range, SDS for Wt and

BMI were significantly lower in the SA group. No significant differences were observed in %FM (SDS), or %FFM (based on original BIA data) in any of the age ranges. However, SA boys had significantly lower SMMa for relative measures of SMMa (i.e. SMMa%; SMMa/FFM%; SMMaI) in all age ranges. MFR (i.e. SMMa (kg)/FM (kg) was significantly lower in SA boys in the 5-7y age range only,

Similar to SA boys, in the 5-7y and 8-10y age ranges, SA girls were significantly shorter, lighter, with a lower BMI for their respective SDS, with no significant differences in %FM (SDS) or %FFM (absolute). In the 11-13y age range no significant differences were observed in Ht, Wt, or BMI SDS, however, SA girls had significantly higher %FM (SDS) and significantly lower %FFM. As with SA boys, compared to their WE_2 counterparts, SA girls in every age range also had significantly lower SMMa for all relative measures (i.e. SMMa%; SMMa/FFM%; SMMaI). MFR was significantly lower in the SA 5-7y and 11-13y age ranges. It is also interesting to note that whilst BMI SDS for both SA boys and girls were significantly lower (except for girls in 11-13y age range – lower but not significantly) than their WE_2 counterparts for all age ranges, %FM SDS were higher (but only significantly higher in the female 11-13y age range) apart from girls in the 8-10y age range (lower but not significantly).

Table 5.2 Descriptive statistics for all BC measures mean ± sd for SA[a] and WE_2[b] cohort with comparisons between ethnic groups by age range and sex

Age & age range (y)	n	Ht SDS	Ht (cm)	Wt SDS	Wt (kg)	BMI SDS	BMI	%FM SDS	FFM (kg)	FFM (%)	SMMa (kg)	SMMa (%)	SMMa/ FFM (%)	MFR	SMMaI
Boys															
[a]6.39 ± 0.93 (5-7)	269 (229)	-0.09** ± 0.97	117.74* ± 7.40	-0.35** ± 1.31	21.40** ± 4.78	-0.48** ± 1.41	15.28** +2.12	0.68 ± 0.87	17.14** ± 3.06	80.81 ± 4.03	5.19** ± 1.57	23.90** ± 2.82	29.73** ± 3.86	1.28** ± 0.28	3.68** ± 0.76
[b]6.40 ± 0.95 (5-7)	247 (246)	0.28 ± 1.05	119.60	0.27 ± 1.35	23.37 ± 5.95	0.11 ± 1.42	16.14 ± 2.57	0.62 ± 0.95	18.67 ± 3.56	80.75 ± 4.74	6.17 ± 1.90	26.10 ± 2.94	32.37 ± 3.93	1.42 ± 0.34	4.22 ± 0.86
[a]9.53 ± 0.88 (8-10)	225 (181)	-0.08** ± 1.04	135.50* ± 8.10	0.04** ± 1.47	32.51* ± 10.18	0.12** ± 1.62	17.41* ± 3.84	0.47 ± 1.17	24.93** ± 5.40	78.51 ± 7.01	8.92** ± 2.69	27.93** ± 2.34	35.64** ± 3.52	1.44 ± 0.44	4.78** ± 0.99
[b]9.45 ± 0.90 (8-10)	206	0.26 ± 0.99	137.05 ± 7.32	0.57 ± 1.27	34.80 ± 10.33	0.62 ± 1.35	18.27 ± 3.93	0.45 ± 1.16	26.72 ± 5.18	78.65 ± 7.03	10.24 ± 2.81	29.59 ± 2.52	37.82 ± 3.21	1.53 ± 0.50	5.37 ± 1.00
[a]12.43 ± 0.84 (11-13)	172 (147)	0.16 ± 0.99	152.75 ± 10.09	0.20* ± 1.24	44.08 ± 11.72	0.07* ± 1.48	18.68 ± 3.59	0.37 ± 1.22	34.45 ± 7.53	79.30 ± 7.13	13.84 ± 3.95	31.16* ± 3.22	39.19** ± 2.73	1.74 ± 0.65	5.72** ± 1.02
[b]12.12 ± 0.72 (11-13)	143	0.35 ± 0.93	151.98 ± 8.39	0.53 ± 0.99	44.52 ± 10.09	0.46 ± 1.11	19.14 ± 3.29	0.32 ± 1.09	35.21 ± 6.41	79.92 ± 5.90	14.30 ± 3.31	32.16 ± 2.90	40.26 ± 2.34	1.76 ± 0.61	6.12 ± 0.94
Girls															
[a]6.4 ± 0.95 5-7	249 (204)	-0.16* ± 1.09	116.86 ± 8.12	-0.50** ± 1.31	20.84** ± 5.14	-0.58** ± 1.33	15.07** ± 2.19	0.44 ± 0.92	16.19** ± 3.31	78.39 ± 3.86	5.08** ± 1.46	24.39** ± 1.85	31.26** ± 2.50	1.15** ± 0.23	3.68** ± 0.62
[b]6.4 ± 0.94 (5-7)	220 (219)	0.14 ± 1.01	118.17 ± 7.67	0.13 ± 1.16	22.65 ± 5.47	0.04 ± 1.21	16.04 ± 2.37	0.43 ± 0.99	17.55 ± 3.40	78.21 ± 4.37	5.88 ± 1.46	25.98 ± 2.21	33.22 ± 2.26	1.24 ± 0.30	4.15 ± 0.58
[a]9.47 ± 0.87 (8-10)	208 (159)	-0.18** ± 1.02	134.35* ± 7.87	-0.32** ± 1.27	30.10** ± 7.23	-0.32** ± 1.27	16.50** ± 2.81	0.03 ± 1.11	22.81** ± 4.28	76.72 ± 5.36	7.83** ± 1.82	26.25** ± 2.04	34.45** ± 1.97	1.17 ± 0.31	4.31** ± 0.64
[b]9.53 ± 0.89 (8-10)	203	0.17 ± 1.06	136.95 ± 8.67	0.36 ± 1.28	34.86 ± 10.48	0.38 ± 1.34	18.28 ± 3.89	0.23 ± 1.14	25.85 ± 5.86	75.60 ± 6.00	9.41 ± 2.53	27.27 ± 2.13	36.12 ± 1.85	1.20 ± 0.35	4.94 ± 0.84
[a]12.17 ± 0.72 (11-13)	458 (456)	0.02 ± 0.95	150.81 ± 7.53	0.11 ± 1.28	43.90 ± 11.01	0.03 ± 1.48	19.15 ± 4.01	0.24* ± 1.18	32.07 ± 5.72	74.37* ± 6.25	11.84* ± 2.46	27.27** ± 2.06	36.73** ± 1.60	1.14* ± 0.35	5.16* ± 0.81
[b]12.08 ± 0.75 (11-13)	118	0.13 ± 0.99	151.06 ± 7.77	0.18 ± 1.13	43.74 ± 10.23	0.09 ± 1.20	19.00 ± 3.47	-0.02 ± 1.00	32.84 ± 6.26	75.95 ± 5.02	12.40 ± 2.72	28.50 ± 2.17	37.56 ± 1.57	1.25 ± 0.32	5.38 ± 0.81

*Significant difference at P< 0.05 level; **Significant difference at P<0.001 level; [a]= South Asians; [b]= White Europeans. Numbers shown in brackets under column 'n' = numbers of subjects with SMMa data available if fewer than total dataset; MFR = Muscle to fat ratio (i.e. SMMa (kg)/ FM (kg)). SMMa = Appendicular skeletal muscle mass; SMMaI = SMMa index (SMMa [kg]/Ht [m^2])
WE_2 = White European low income dataset

Further analyses on absolute and relative FFM (kg & % respectively) were conducted to assess the differences if any between the SA and WE_2 cohort before and after regression following validation with DXA (Chapter 4). Data is shown in Table 5.3 and is limited to ages ≥9y as explained in the introduction. Application of BIA_{reg1} to the SA sample resulted in a small within-group decrease in both absolute and relative FFM for both boys (mean difference = 0.35kg; & 1.15% P <0.001) and girls (mean difference =0.10kg; & 0.29%), although for girls the decrease in FFM was not significant. Both SA boys and girls had less relative %FFM than their WE_2 counterparts. Between-group comparisons (WE_2 – SA) for boys revealed no significant difference between SA and WE_2 groups in FFM% before BIA correction (mean difference = 0.67%; SE = 0.64), however, after correction the difference was significant (mean difference = 1.82%; SE = 0.70; P = 0.009). For girls, the average difference in %FFM was not significant before (mean difference = 0.44%; SE = 0.45) or after (mean difference = 0.73%; SE = 0.81) correction.

Table 5.3 Descriptive statistics (mean ± sd) for SA & WE_2 absolute (kg) and relative FFM (%) showing sex-specific comparisons before and after regression between ethnic groups aged 9-14y

	Age (y)	**n**	**FFM (kg)**	**FFM (%)**	**FFM_{reg1} (kg)**	**FFM_{reg1} (%)**
SA boys	11.29 ± 1.42	322	30.64* ± 7.90	78.42 ± 8.55	30.29* ± 8.47	77.26* ± 9.62
WE_2 boys	11.14 ± 1.23	271	31.94 ± 6.86	79.08 ± 6.84		
SA girls	11.68 ± 1.14	593	30.32 ± 6.38	74.92 ± 6.19	30.22 ± 7.90	74.63 ± 12.41
WE_2 girls	10.98 ± 1.23	255	30.02 ± 6.56	75.35 ± 5.73		

*Significant difference at P< 0.05 level; SA = South Asian; WE_2 = White European low income dataset

FFM = fat free mass; FFM_{reg1} = FFM regression equation 1 (see section 4.5.3.5)

Tables 5.4a and 5.4b show descriptive statistics for SA (boys and girls respectively) sub-groups (i.e. Indian, Pakistani, Bangladeshi, and other Asian), and Table 5.5 shows comparisons, where differences were significant (P< 0.05 or 0.001) within-

group and between-group (i.e. comparisons between SA sub-groups and the WE_2 cohort presented in Table 5.1). Significant differences between SA sub-groups and WE_2 boys and girls were observed across all measures apart from %FM, where there were no significant differences.

Compared to WE_2 boys, Indian boys, were significantly shorter (Ht SDS; 5-7y age range only), and lighter (Wt SDS), with a lower BMI (SDS) in the 5-7y and 8-10y age ranges, with no significant differences for these measures in the 11-13y age range. In the 8-10y age range Indian boys were also significantly lighter than Pakistani boys, with a significantly lower BMI than both Pakistani and Bangladeshi boys, and also had significantly less %FM than Pakistani boys. For all SMMa measures, Indian boys had significantly less SMMa than WE_2 boys in the 5-7y and 8-10y age ranges, with less relative SMMa (i.e. SMMa%; SMMa/FFM%; SMMaI) in the 11-13y age range. They also had less SMMa (SMMa%, SMMa/FFM%, and SMMaI) than Bangladeshi boys in the 5-7y and 8-10y (SMMaI only) age ranges, as well as less SMMa (SMMa/FFM% only) than Pakistani boys in the 5-7y age range.

Bangladeshi boys in one or more age ranges, compared to WE_2 boys, were significantly shorter (in all three age ranges) and lighter (in 5-7y and 8-10y age ranges), with no significant differences in BMI. Bangladeshi boys also had significantly less SMMa (SMMa% and SMMa/FFM %) than WE_2 boys in the 8-10y age range. Pakistani boys had significantly less SMMa (%) than WE_2 boys in the 8-10y age range only, with no significant differences for any other measures.

For the female dataset, compared to WE_2 girls, Indian girls were significantly shorter (8-10y age range only), lighter, with a lower BMI in the 5-7y and 8-10y age ranges. Some or all measures of SMMa were also significantly lower in Indian girls across all age ranges compared to WE_2 girls. Indian girls also had significantly less SMMa (SMMa/FFM% and SMMaI) than Bangladeshi girls in the 5-7y and 8-10y age ranges. Compared to WE_2 girls, Pakistani girls were significantly lighter (5-7y age range), with no other significant differences across all age ranges for Ht, Wt or BMI measures. For SMMa measures Pakistani girls had significantly less SMMa compared with WE_2 girls, in the 5-7y (SMMa/FFM%; SMMaI), and 11-13y (SMMa%;

SMMa/FFM%; MFR) age ranges. There were no sub-group differences between Pakistani girls across all age ranges for any measures.

Bangladeshi girls, compared to WE_2 girls, were significantly lighter (11-13y age range), with a lower BMI (8-10y age range). Bangladeshi girls had significantly lower SMMa for one or more measures across all three age ranges compared to WE_2 girls.

Table 5.4a Descriptive statistics for all BC measures (mean ± sd) for SA sub-groups by age range (boys only)

Age (y) & age range α, β, γ	n	Ht SDS	Ht (cm)	Wt SDS	Wt (kg)	BMI SDS	BMI	WC SDS	%FM SDS	FFM (kg)	FFM (%)	SMMa (kg)	SMMa (%)	SMMa/ FFM (%)	MFR	SMMaI
1α6.19 ± 0.93	126	-0.08 ± 1.02	116.55 ± 7.61	-0.47 ± 1.28	20.55 ± 4.34	-0.66 ± 1.31	15.00 ± 1.92	-0.28 ± 1.19	0.73 ± 0.79	16.52 ± 2.77	80.92 ± 3.70	4.87 ± 1.40	23.40 ± 2.79	28.97 ± 3.75	1.26 ± 0.26	3.52 ± 0.69
2α6.48 ± 0.85	37	0.13 ± 0.76	119.46 ± 6.54	0.05 ± 1.19	22.68 ± 4.78	-0.10 ± 1.31	15.75 ± 2.04	0.52 ± 1.29	0.85 ± 0.90	17.97 ± 2.96	79.89 ± 4.34	5.65 ± 1.52	24.66 ± 2.39	30.94 ± 3.35	1.28 ± 0.30	3.89 ± 0.70
3α6.70 ± 0.86	91 (53)	-0.20 ± 0.97	119.04 ± 7.06	-0.33 ±1.42	22.26 ±5.24	-0.35 ±1.56	15.55 ±2.42	0.14 ±1.29	0.55 ±0.98	17.81 ±3.31	80.85 ±4.46	5.78 ±1.77	24.90 ±2.74	31.19 ±3.77	1.31 ±0.31	3.98 ±0.85
4α5.96 ±0.94	15 (13)	0.02 ± 0.91	115.67 ±7.91	-0.44 ±1.07	20.07 ±4.14	-0.75 ±1.26	14.85 ±1.51	-0.57 ±1.19	0.62 ±0.65	16.35 ±3.07	81.70 ±2.38	4.50 ±1.39	22.62 ±3.11	27.76 ±4.13	1.25 ±0.22	3.38 ±0.71
1β9.44 ±0.84	84 (82)	-0.09 ±1.10	134.96 ±8.04	-0.28 ±1.42	30.19 ±8.63	-0.36 ±1.64	16.36 ±3.23	0.20 ±1.42	0.19 ±1.23	23.66 ±4.57	79.83 ±6.88	8.46 ±2.50	27.87 ±2.46	35.07 ±3.61	1.52 ±0.45	4.55 ±0.92
2β9.80 ±0.78	36 (21)	0.27 ±1.00	138.96 ±7.12	0.56 ±1.36	36.33 ±11.11	0.60 ±1.50	18.52 ±4.03	1.41 ±1.15	0.87 ±1.00	27.11 ±5.74	76.40 ±7.03	9.18 ±2.23	27.65 ±1.64	36.04 ±2.61	1.30 ±0.36	4.82 ±0.80
3β9.52 ±0.91	90 (63)	-0.26** ±1.01	134.41 ±8.27	0.06 ±1.56	32.96 ±11.03	0.30 ±1.62	17.89 ±4.26	0.59 ±1.48	0.52 ±1.19	25.04 ±5.77	78.11 ±7.24	9.27 ±3.01	27.96 ±2.23	36.10 ±3.68	1.39 ±0.44	5.00 ±1.10
4β9.42 ±1.06	15	0.22 (0.79	136.74 ±7.85	0.53 ±1.02	33.62 ±7.83	0.59 ±1.11	17.73 ±2.49	0.92 ±0.86	0.72 ±0.84	26.12 ±5.06	78.37 ±5.02	9.61 ±2.69	28.44 ±3.05	36.29 ±3.22	1.39 ±0.41	5.06 ±0.89
1γ12.61 ±0.82	71	0.24 ±1.05	154.51 ±10.32	0.21 ±1.40	45.62 ±13.78	0.00 ±1.61	18.78 ±3.88	0.45 ±1.33	0.45 ±1.31	35.22 ±8.76	78.59 ±7.86	14.01 ±4.47	30.70 ±3.41	39.13 ±2.95	1.67 ±0.69	5.73 ±1.16
2γ12.30 ±0.86	42 (31)	0.27 ±1.06	152.08 ±10.31	0.32 ±1.23	44.29 ±10.84	0.15 ±1.55	18.84 ±3.64	0.59 ±1.37	0.46 ±1.20	34.49 ±6.93	78.90 ±6.72	14.07 ±3.55	31.34 ±2.96	39.27 ±2.51	1.74 ±0.66	5.76 ±0.94
3γ12.07 ±0.76	41 (27)	-0.21 ±0.81	147.77 ±8.49	0.08 ±1.14	40.99 ±9.57	0.20 ±1.33	18.66 ±3.56	0.37 ±1.25	0.30 ±1.15	32.30 ±6.32	79.70 ±6.80	12.75 ±3.62	31.25 ±3.23	38.93 ±2.88	1.77 ±0.57	5.61 ±0.97
4γ12.83 ±0.78	18	0.40 ±0.84	157.07 ±8.15	0.16 ±0.85	44.57 ±8.21	-0.16 ±1.02	17.96 ±2.34	0.01 ±0.92	-0.01 ±1.02	36.27 ±5.28	82.02 ±5.41	14.45 ±2.60	32.52 ±2.65	39.67 ±2.04	1.96 ±0.57	5.82 ±0.68

BC = body composition; SA = South Asian; Ht = height; Wt= weight; BMI= body mass index; WC = waist circumference; FM = fat mass; FFM = fat-free mass; SDS = standard deviation scores; numbers 1 to 5 under column 'Age & age range' = ethnic codes where 1 = Indian; 2 = Pakistani; 3 = Bangladeshi; 4 = Other Asian; α, β, γ under column 'Age & age range' = age ranges 5 to 7y; 8 to10y; & 11to13y respectively. Numbers shown in brackets under column 'N' = numbers of subjects with SMMa data available if fewer than total dataset, including MFR. MFR = Muscle to fat ratio (i.e. SMMa (kg)/ FM (kg)). SMMa = Appendicular skeletal muscle mass; SMMaI = SMMa index (SMMa [kg]/Ht [m^2])

Table 5.4b Descriptive statistics for all BC measures (mean ± sd) for SA sub-groups by age range (girls only)

Age (y) & age range α, β, γ	n	Ht SDS	Ht (cm)	Wt SDS	Wt (kg)	BMI SDS	BMI	WC SDS	%FM SDS	FFM (kg)	FFM (%)	SMMa (kg)	SMMa (%)	SMMa/ FFM (%)	MFR	SMMaI
1α6.23 ±1.00	116	-0.08 ±1.07	116.35 ±8.79	-0.55 ±1.31	20.45 ±5.42	-0.73 ±1.37	14.88 ±2.24	-0.36 ±1.35	0.54 ±0.91	15.84 ±3.48	78.19 ±3.99	4.99 ±1.46	24.24 ±1.82	31.03 ±2.45	1.15 ±0.24	3.61 ±0.61
2α6.49 ±0.97	15 (14)	-0.47 ±0.95	115.81 ±6.67	-0.82 ±1.23	19.82 ±4.06	-0.78 ±1.19	14.66 ±1.77	-0.31 ±1.21	0.13 ±0.86	15.73 ±2.81	79.67 ±2.82	4.89 ±1.16	24.98 ±1.76	31.42 ±2.25	1.24 ±0.21	3.65 ±0.56
3α6.57 ±0.87	88 (45)	-0.14 ±1.14	117.95 ±7.68	-0.33 ±1.27	21.58 ±4.69	-0.35 ±1.23	15.38 ±2.16	0.14 ±1.17	0.33 ±0.96	16.82 ±2.99	78.54 ±3.86	5.53 ±1.45	24.95 ±1.62	32.20 ±2.13	1.15 ±0.23	3.94 ±0.60
4α6.42 ±0.97	30 (29)	-0.32 ±1.09	116.12 ±8.09	-0.63 ±1.46	20.70 ±5.71	-0.63 ±1.52	15.07 ±2.22	-0.26 ±1.41	0.58 ±0.85	15.96 ±3.65	77.91 ±3.73	4.90 ±1.49	23.88 ±2.13	30.62 ±3.05	1.12 ±0.20	3.58 ±0.67
1β9.37 ±0.87	90 (88)	-0.28 ±1.07	133.22 ±7.79	-0.51 ±1.22	28.73 ±6.56	-0.50 ±1.20	16.04 ±2.59	-0.02 ±1.26	0.07 ±1.01	21.81 ±3.86	76.72 ±5.08	7.48 ±1.73	26.13 ±2.09	34.08 ±2.10	1.18 ±0.30	4.17 ±0.60
2β9.73 ±0.91	14	0.05 ±1.11	137.34 ±9.19	-0.10 ±1.41	32.37 ±8.40	-0.21 ±1.60	16.96 ±3.08	0.52 ±1.51	0.07 ±1.27	24.37 ±5.04	76.29 ±5.55	8.61 ±2.13	26.71 ±1.98	35.03 ±1.81	1.20 ±0.37	4.50 ±0.69
3β9.50 ±0.82	89 (42)	-0.14 ±0.98	134.85 ±7.84	-0.20 ±1.31	30.99 ±7.70	-0.19 ±1.37	16.85 ±3.00	0.28 ±1.41	-0.07 ±1.21	23.52 ±4.53	76.96 ±5.88	8.27 ±1.91	26.50 ±2.11	35.12 ±1.78	1.17 ±0.36	4.56 ±0.68
4β9.59 ±1.03	15	-0.10 ±0.94	135.43 ±6.64	-0.15 ±1.10	30.95 ±6.14	-0.15 ±1.10	16.77 ±2.55	0.45 ±1.20	0.30 ±0.88	23.21 ±3.49	75.74 ±4.45	7.96 ±1.41	25.85 ±1.51	34.17 ±1.28	1.11 ±0.24	4.31 ±0.53
1γ12.38 ±0.74	168	0.11 ±0.88	152.59 ±7.31	0.08 ±1.19	43.38 ±10.33	-0.07 ±1.41	18.95 ±3.73	0.31 ±1.50	0.23 ±1.13	32.57 ±5.50	74.47 ±5.84	12.00 ±2.37	27.28 ±1.97	36.67 ±1.42	1.14 ±0.34	5.12 ±0.78
2γ12.08 ±0.67	143	0.06 ±1.04	150.62 ±7.59	0.23 ±1.38	44.63 ±11.53	0.16 ±1.56	19.50 ±4.22	0.63 ±1.59	0.35 ±1.24	32.26 ±6.00	73.73 ±6.71	11.95 ±2.66	27.08 ±2.14	36.82 ±1.83	1.11 ±0.36	5.22 ±0.89
3γ11.93 ±0.62	119 (107)	-0.08 ±0.93	148.79 ±7.13	0.11 ±1.31	42.89 ±11.81	0.09 ±1.49	19.17 ±4.29	0.34 ±1.55	0.18 ±1.23	31.35 ±5.88	74.67 ±6.47	11.56 ±2.41	27.42 ±2.11	36.77 ±1.56	1.16 ±0.35	5.16 ±0.79
4γ12.38 ±0.79	28	-0.30 ±0.86	149.70 ±8.07	-0.24 ±1.13	41.56 ±8.35	-0.20 ±1.35	18.49 ±3.29	0.21 ±1.40	0.03 ±1.02	31.11 ±4.70	75.65 ±5.19	11.41 ±2.02	27.61 ±1.99	36.52 ±1.48	1.19 ±0.31	5.07 ±0.71

BC = body composition; SA = South Asian; Ht = height; Wt= weight; BMI= body mass index; WC = waist circumference; FM = fat mass; FFM = fat-free mass; SDS = standard deviation scores; numbers 1 to 5 under column 'Age & age range' = ethnic codes where 1 = Indian; 2 = Pakistani; 3 = Bangladeshi; 4 = Other Asian

α, β, γ under column 'Age & age range' = age ranges 5 to 7y; 8 to10y; & 11to13y respectively.

Numbers shown in brackets under column 'N' = numbers of subjects with SMMa data available if fewer than total dataset, including MFR. MFR = Muscle to fat ratio (i.e. SMMa (kg)/ FM (kg)).

SMMa = Appendicular skeletal muscle mass; SMMaI = SMMa index (SMMa [kg]/Ht [m^2])

Table 5.5 Descriptive statistics showing significant (* or **) differences within SA sub-groups and between SA sub-groups & WE_2 for all BC measures by sex and age range

	Ethnic groups (1to 5) with significant differences (mean difference)					
Variables	**Boys**			**Girls**		
Age ranges (y)	**5 to 7**	**8 to 10**	**11 to13**	**5 to 7**	**8 to10**	**11to 13**
Ht SDS	1 - 5 (-0.37) * 3 - 5 (-0.49) **	3 – 5 (-0.52) **	3 – 5 (-0.56) *	-	1 – 5 (-0.33) *	-
Ht (cm)	1 – 5 (-3.04) *	2 – 3 (4.55) *	1 – 3 (6.73) * 3 – 4 (-9.30) *	-	1 – 5 (-3.72) *	1 – 3 (3.80) **
Wt SDS	1 - 5 (-0.74) ** 3 - 5 (-0.60) *	1 – 2 (-0.84) * 1 – 5 (-0.85) ** 3 – 5 (-0.50) *	-	1 -5 (-0.68) ** 2 – 5 (-0.95) * 3 -5 (-0.45) * 4 -5 (-0.76) *	1 – 5 (-0.86) ** 3 – 5 (-0.55) *	-
Wt (kg)	1 – 5 (-2.82) **	1 – 2 (-6.14) * 1 – 5 (4.61) *	-	1 – 5 (-2.19) *	1 – 5 (-6.13) ** 3 – 5 (-3.87) *	-
BMI SDS	1 - 5 (-0.76) **	1 – 2 (-0.95) * 1 – 3 (-0.65) * 1 – 5 (-0.97) **	-	1 – 5 (-0.77) **	1 – 5 (-0.88) ** 3 – 5 (-0.56) *	-
BMI	1 – 5 (-1.14) **	1 – 2 (-2.16) * 1 – 5 (-1.91) **	-	1 – 5 (-1.15) **	1 – 5 (-2.24) ** 3 – 5 (-1.43) *	-
WC SDS	1 - 2 (-0.79) * 1 - 5 (-0.93) ** 2 - 4 (1.09) * 3 - 5 (-0.52) * 4 -5 (-1.23) *	1 – 2 (-1.21) ** 1 - 5 (-0.64) * 2 - 3 (0.82) *	-	1 - 3 (-0.50) * 1 - 5 (-0.88) ** 4 – 5 (-0.78) *	1 – 5 (-0.78) ** 3 – 5 (-0.48) *	-
%FM SDS	-	1 - 2 (-0.67) *	-	-	-	-
FFM (kg)	1 – 3 (-1.29) * 1 – 5 (-2.15) **	1 – 2 (-3.45) * 1 – 5 (-3.07) **	-	1 – 5 (-1.71) **	1 – 5 (-4.03) ** 3 – 5 (-2.33) *	-
FFM (%)	-	-	-	-	-	2 – 5 (-2.21) *
SMMa (kg)	1 - 3 (-0.92) * 1 - 5 (-1.30) ** 4 - 5 (-1.67) *	1 – 5 (-1.78) **	-	1 – 5 (-0.90) ** 4 – 5 (-0.98) *	1 -5 (-1.93) ** 3 – 5 (-1.14) *	-
SMMa (%)	1 - 3 (-1.51) * 1 - 5 (-2.68) ** 2 - 5 (-1.42) * 4 - 5 (-2.28) **	1 – 5 (-1.72) ** 2 – 5 (-1.94) * 3 – 5 (-1.62) **	1 – 5 (-1.46) *	1 – 5 (-1.74) ** 3 – 5 (-1.03) * 4 – 5 (-2.10) **	1 – 5 (-1.14) **	1 – 5 (-1.22) ** 2 – 5 (-1.42) ** 3 – 5 (-1.08) **
SMMa/ FFM (%)	1 - 2 (-1.97) * 1 - 3 (-2.22) * 1 – 5 (-3.40) ** 3 – 4 (3.43) * 4 – 5 (-4.61) **	1 – 5 (-2.76) ** 3 – 5 (-1.73) *	1 – 5 (-1.13) *	1 – 3 (-1.16) * 1 – 5 (-2.19) ** 2 – 5 (-1.81) * 3 - 4 (1.58) * 4 – 5 (-2.60) **	1 – 3 (-1.03) * 1 – 5 (-2.03) ** 3 – 5 (-1.00) * 4 – 5 (-1.94) **	1 – 5 (-0.89) ** 2 – 5 (-0.74) * 3 – 5 (-0.79) * 4 – 5 (1.03) *
MFR	1 – 5 (-0.16) **	-	-	1 – 5 (-0.10) *	-	2 – 5 (-0.13) *
SMMaI	1 – 3 (-0.46) * 1 – 5 (-0.70) ** 4 – 5 (-0.84) *	1 – 3 (-0.45) * 1 – 5 (-0.82) **	1 – 5 (-0.39) *	1 – 3 (-0.33) * 1 – 5 (-0.54) ** 2 – 5 (-0.50) * 4 – 5 (-0.57) **	1 – 3 (-0.40) * 1 – 5 (-0.77) **	-

SA = South Asian; WE_2 = White European low income dataset; ethnic groups: 1 = Indian; 2 = Pakistani; 3 = Bangladeshi; 4 = Other Asian; 5 = White European

*Significant difference at P< 0.05 level; **Significant difference at P<0.001 level;

Data presented beneath each age-range column shows which two groups are significantly different, where '- 'represents minus or subtraction e.g. 1-5 indicates significant differences between ethnic groups 1 & 5, where mean difference in brackets = mean value of group1 minus mean value of group 5 for the indicated variable in column 1.

Ht = height; Wt = weight; BMI = body mass index; WC = waist circumference; FM = fat mass; FFM = fat-free mass; SMM_a = appendicular skeletal muscle mass; MFR = muscle-to-fat ratio (SMMa [kg]/ FM [kg]); SMMaI = SMMa index (SMMa [kg]/Ht [m^2])

Tables 5.6 – 5.7 and 5.8 – 5.9 are % FM centile values for SA boys and girls respectively based on the original BIA output (Tables 5.6 & 5.8) for Tanita (BC418-MA) and the corrected output (Tables 5.7 & 5.9) using the new regression equation developed in the BIA validation study (chapter 4 section 4.5.3.4). Due to too few participants < 9y, the new regression equations were only applicable to SA children and adolescents ≥ 9y. Centile curves for %FM are presented in Figures 5.1a and 5.2a (original equations) and 5.1b and 5.2b (new regression equations) for boys and girls respectively following application of the new equations to the existing SA dataset. Both centile charts and curves before and after correction are presented for comparative purposes. Due to very few subjects < 14y, the %FM centiles for 9-14y, for both boys and girls are presented.

BIA prior to correction tended to overestimate %FM at the leaner end of the spectrum (≤ 15%FM in boys and ≤ 20% FM in girls) and underestimate %FM at high adiposity levels for both boys and girls (Tables 5.6 and 5.8 and Figures 5.1a and 5.2a for boys and girls respectively). For boys, the new regression equation (Table 5.7, and Figure 5.1b) appeared to compensate for %FM underestimation at the 2nd and 9th centiles particularly with increasing age; this was also observed at the 25th centile in ages ≥12y. At the 50th and above centiles, higher %FM values were observed overall with the new regression equations, particularly in the 9-11y ages, with an approximate increase in %FM of 3%.

For girls, similar to boys, the centiles produced from the BIA regression equation (Table 5.9 and Figure 5.2b) resulted in lower %FM values in the 2nd and 9th centiles and higher %FM values above the 50th centile, than the values computed from the original BIA equation (Table 5.8 and Figure 5.2a). This result indicates that the new regression equation was able to correct for BIA overestimation of %FM at the leaner end of the scale, and underestimation of %FM at higher adiposity levels.

Table 5.6. %Fat centiles by age based on original BIA output (SA Boys)

BIA original		**Centiles**								
		-2.05	-1.34	-0.67	0	0.67	1.04	1.34	1.64	2.05
Age (y)	**n**	2	9	25	50	75	85	91	95	98
9	39	12.73	14.51	16.68	19.65	23.91	27.04	30.38	34.66	42.73
9.5	29	12.64	14.55	16.89	20.09	24.65	27.99	31.52	35.98	44.20
10	46	12.49	14.53	17.03	20.45	25.28	28.78	32.45	37.01	45.24
10.5	38	12.24	14.38	16.99	20.56	25.57	29.16	32.89	37.47	45.56
11	32	11.95	14.13	16.79	20.42	25.50	29.13	32.86	37.42	45.37
11.5	35	11.70	13.85	16.51	20.13	25.19	28.80	32.52	37.07	44.99
12	19	11.44	13.54	16.12	19.65	24.61	28.15	31.82	36.31	44.17
12.5	31	11.19	13.20	15.68	19.07	23.84	27.27	30.82	35.18	42.87
13	42	10.90	12.82	15.18	18.39	22.90	26.13	29.46	33.56	40.76
13.5	13	10.66	12.50	14.75	17.78	21.98	24.94	27.98	31.67	38.05

BIA = Bioelectrical impedance analysis; SA = South Asian

Table 5.7. %Fat centiles by age based on BIA_{reg1} (SA Boys)

	Centiles									
Age (y)	**n**	2	9	25	50	75	85	91	95	98
9	39	11.34	14.82	18.58	22.89	27.74	30.56	33.06	35.67	39.37
9.5	29	10.65	14.50	18.65	23.41	28.72	31.81	34.53	37.37	41.36
10	46	9.84	14.05	18.60	23.79	29.55	32.88	35.80	38.83	43.08
10.5	38	8.80	13.32	18.20	23.73	29.84	33.34	36.40	39.57	43.99
11	32	7.66	12.39	17.49	23.27	29.60	33.22	36.37	39.62	44.14
11.5	35	6.63	11.45	16.67	22.59	29.08	32.77	36.00	39.31	43.93
12	19	5.81	10.53	15.72	21.64	28.16	31.89	35.15	38.51	43.18
12.5	31	5.25	9.73	14.74	20.54	27.00	30.72	33.98	37.35	42.07
13	42	4.89	9.04	13.78	19.34	25.61	29.25	32.46	35.80	40.49
13.5	13	4.61	8.41	12.81	18.02	23.95	27.43	30.50	33.71	38.24

BIA_{reg1} = Bioelectrical impedance analysis regression equation 1 (see section 4.5.3.5); SA = South Asian

Table 5.8 %Fat centiles by age based on original BIA output (SA Girls)

Centiles										
		-2.05	-1.34	-0.67	0	0.67	1.04	1.34	1.64	2.05
Age	**n**	2	9	25	50	75	85	91	95	98
9	31	15.72	17.34	19.26	21.77	25.21	27.63	30.13	33.22	38.78
9.5	38	15.46	17.21	19.26	21.93	25.50	27.98	30.49	33.52	38.74
10	35	15.22	17.11	19.30	22.14	25.86	28.38	30.88	33.82	38.71
10.5	30	15.08	17.12	19.49	22.50	26.37	28.93	31.43	34.30	38.91
11	104	15.14	17.37	19.93	23.14	27.19	29.81	32.31	35.14	39.57
11.5	104	15.43	17.86	20.63	24.05	28.27	30.96	33.49	36.30	40.60
12	87	15.76	18.38	21.33	24.93	29.29	32.01	34.53	37.31	41.48
12.5	103	15.90	18.64	21.70	25.37	29.75	32.44	34.92	37.61	41.60
13	49	15.92	18.74	21.84	25.52	29.85	32.48	34.87	37.45	41.23
13.5	11	15.93	18.81	21.94	25.62	29.89	32.45	34.76	37.23	40.82

BIA = Bioelectrical impedance analysis; SA = South Asian

Table 5.9 %Fat centiles by age based on BIA_{reg1} (SA Girls)

		Centiles								
		-2.05	-1.340	-0.67	0	0.67	1.04	1.34	1.64	2.05
Age	**N**	2	9	25	50	75	85	91	95	98
9	31	11.05	15.15	19.49	24.36	29.70	32.76	35.43	38.20	42.05
9.5	38	10.65	15.01	19.52	24.47	29.77	32.76	35.35	38.00	41.66
10	35	10.24	14.89	19.57	24.58	29.85	32.77	35.28	37.82	41.30
10.5	30	9.90	14.83	19.67	24.75	29.98	32.84	35.28	37.73	41.07
11	104	9.78	14.96	19.94	25.07	30.29	33.12	35.51	37.92	41.17
11.5	104	9.99	15.33	20.40	25.58	30.81	33.63	36.01	38.39	41.61
12	87	10.44	15.83	20.92	26.11	31.33	34.14	36.51	38.88	42.08
12.5	103	10.97	16.25	21.24	26.33	31.46	34.22	36.56	38.89	42.03
13	49	11.56	16.62	21.43	26.35	31.33	34.02	36.29	38.56	41.63
13.5	11	12.29	17.08	21.67	26.41	31.23	33.84	36.05	38.27	41.28

BIA_{reg1} = Bioelectrical impedance analysis regression equation 1 (see section 4.5.3.5); SA = South Asian

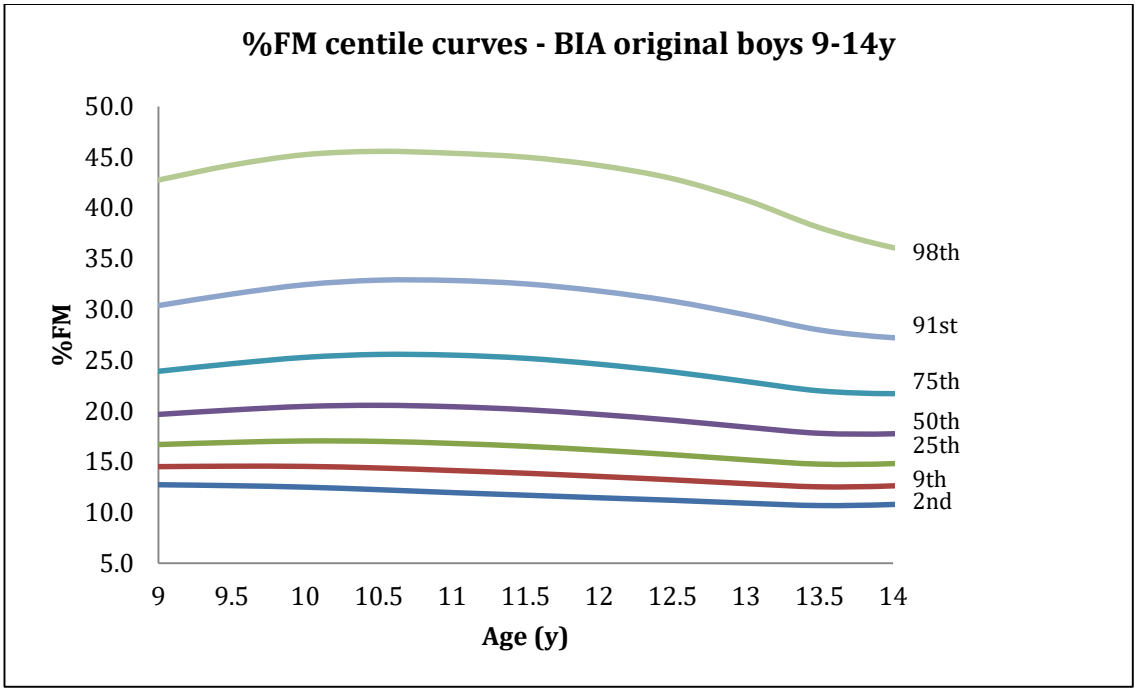


Figure 5.1a % FM centiles (original BIA equation) SA boys. FM = fat mass; BIA = bioelectrical impedance analysis; SA = South Asian

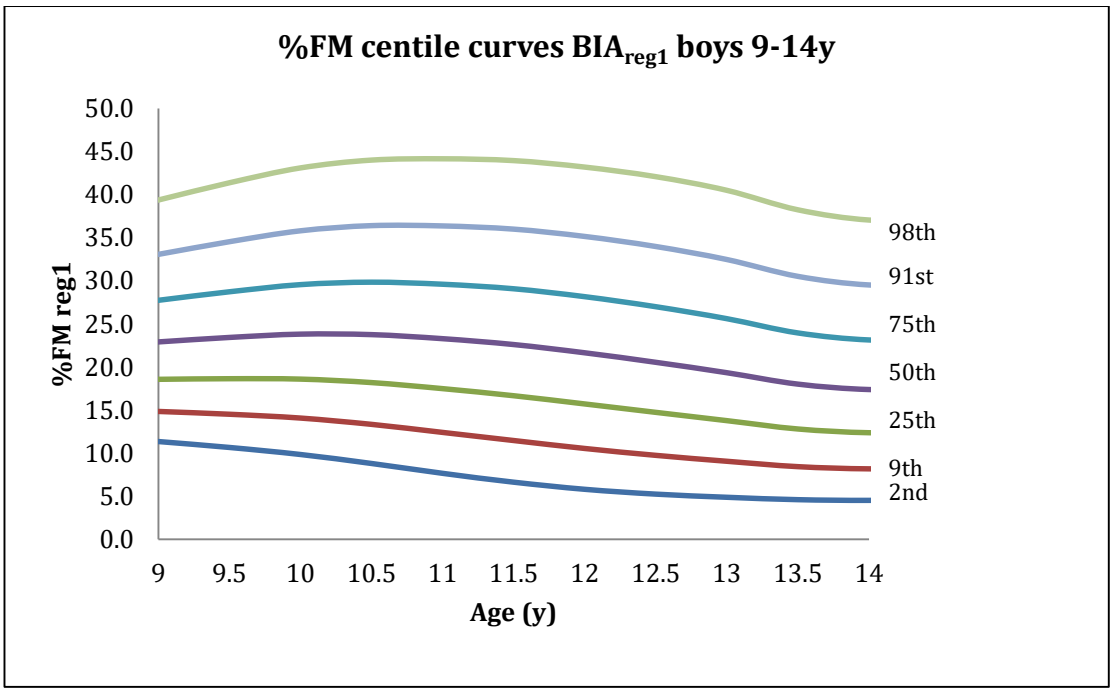


Figure 5.1b - %FM centiles SA boys (new BIAreg1 equation developed see section 4.5.3.5). FM = fat mass; BIA = bioelectrical impedance analysis; SA = South Asian

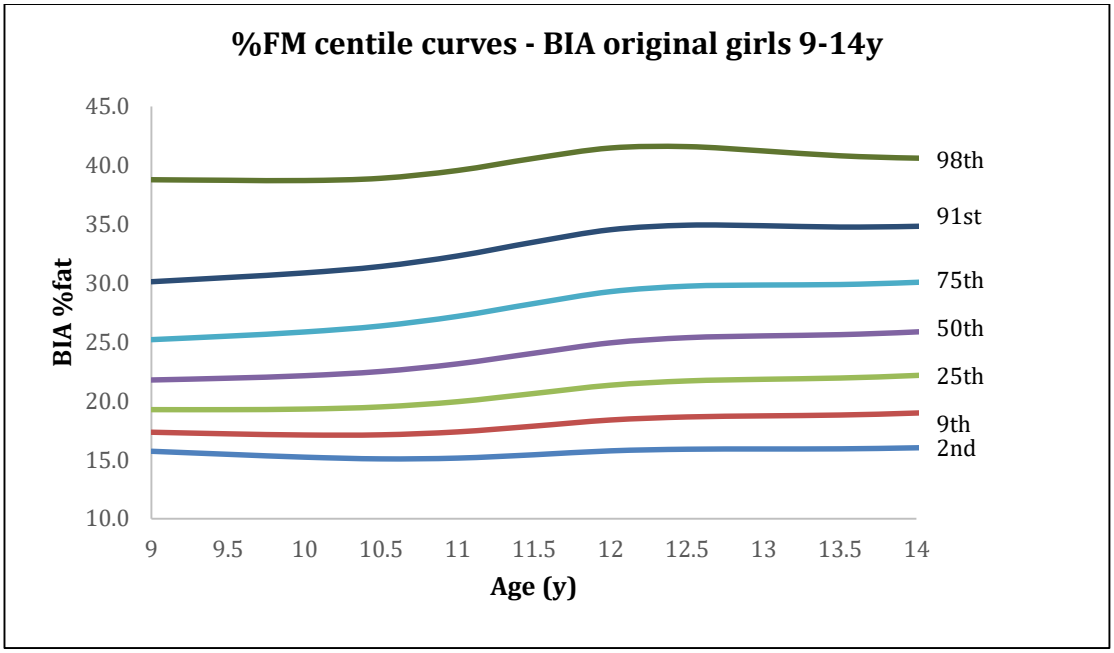


Figure 5.2a % FM centiles SA girls (original BIA equation). FM = fat mass; BIA = bioelectrical impedance analysis; SA = South Asian

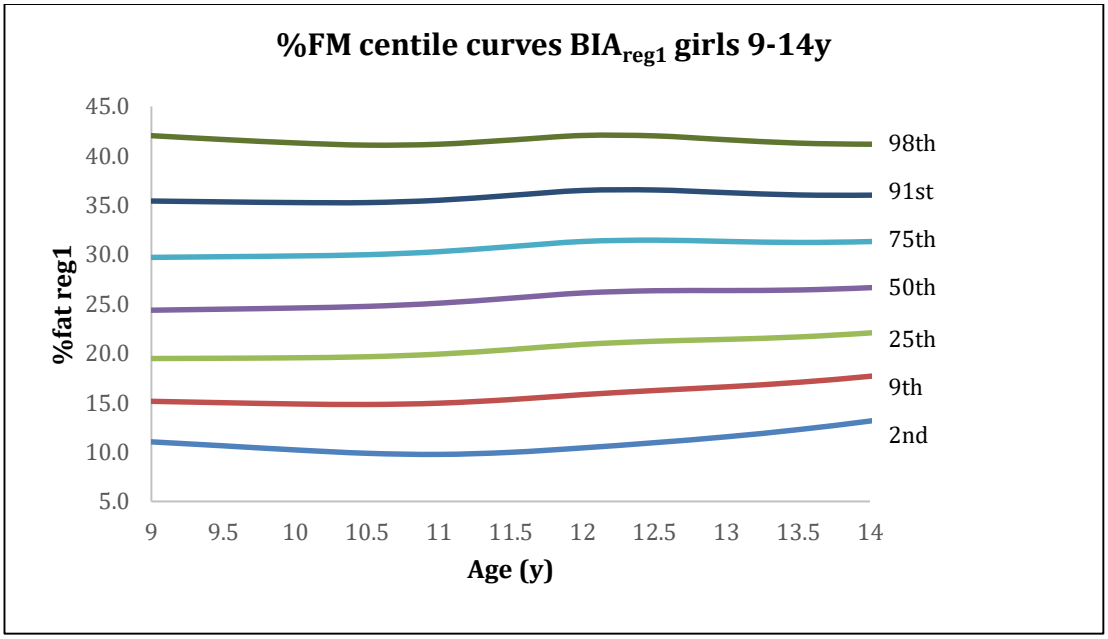


Figure 5.2b %FM centiles SA girls (new BIAreg1 equation developed see section 4.5.3.5). FM = fat mass; BIA = bioelectrical impedance analysis; SA = South Asian

Figures 5.3a (boys) and 5.3b (girls) show sex-specific BIA %FM comparisons between WE_1 (McCarthy et al., 2006), and SA (BIA original and new regression

equation) datasets at the 50th centile. For SA boys, the new BIA regression equation compared to the proprietary inbuilt equation, resulted in higher %FM values up until age 13.5y where the values appeared to converge, with the largest differences observed in ages <11y. It should be noted however, that there were only 11 subjects in the 13.5 -14y age group. Compared to WE_1 boys, SA boys had higher %FM at the 50th centile, with the greatest differences observed in children below age11y.

For SA girls, similar to boys, the new BIA regression equation resulted in higher %FM values across all ages, with the largest differences observed in children <11.5y. Compared to WE_1 girls, the original BIA equation indicated that SA girls < 11y had less %FM than WE_1 girls, and >11y had higher %FM. However, the new BIA regression equation indicated that SA girls had higher %FM than WE_1 girls across all ages.

It is important to note that whilst no significant differences were observed between SA and WE_2 children for %FM SDS or %FFM (apart from girls aged 11-13y) by age range as reported in Tables 5.1-5.3 above, the WE_2 dataset selected was from children from low-income backgrounds, to ensure that socioeconomic status did not influence the results. In contrast, the WE_1 children involved in the study conducted by McCarthy et al. (2006) were intentionally selected from more affluent backgrounds to minimise the effect on the outcome of rising levels of obesity in order to make comparisons with the UK90 reference population.

As %FM comparisons at the 50th centile were made between the WE_1 dataset (between ages 9-14y) from the study conducted by McCarthy et al. (2006) and SA dataset using both the BIA original equation and the new BIA_{reg1} equation developed in chapter 4, similar comparisons were made with %FFM data using the reciprocal values for %FM (Figures 5.4a & 5.4b) as well as at the 2nd, 50th, and 98th centiles (Figures 5.5a & 5.5b). The WE_2 low-income dataset was also included in the comparison to illustrate the differences between the WE_1 sample used in the McCarthy et al. (2006) study.

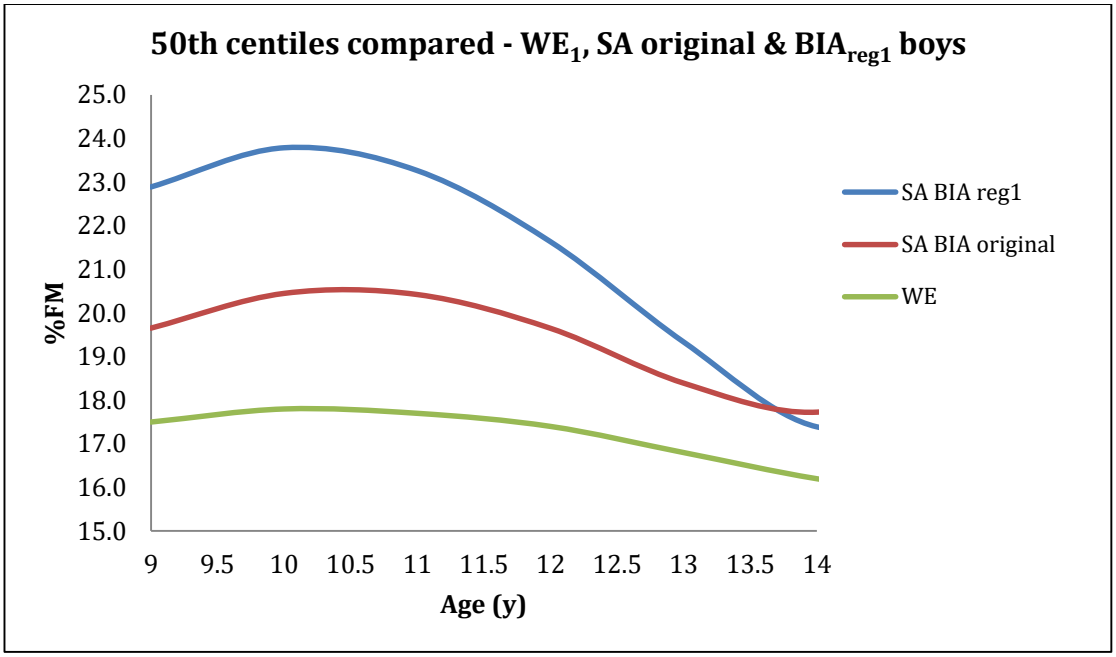


Figure 5.3a %FM comparisons between WE_1 and SA boys before and after application of new BIAreg1 (BIA regression equation see section 4.5.3.5); SA = South Asian; WE_1 = White European data from McCarthy et al. (2006) study. Fat mass = fat mass; BIA =bioelectrical impedance analysis

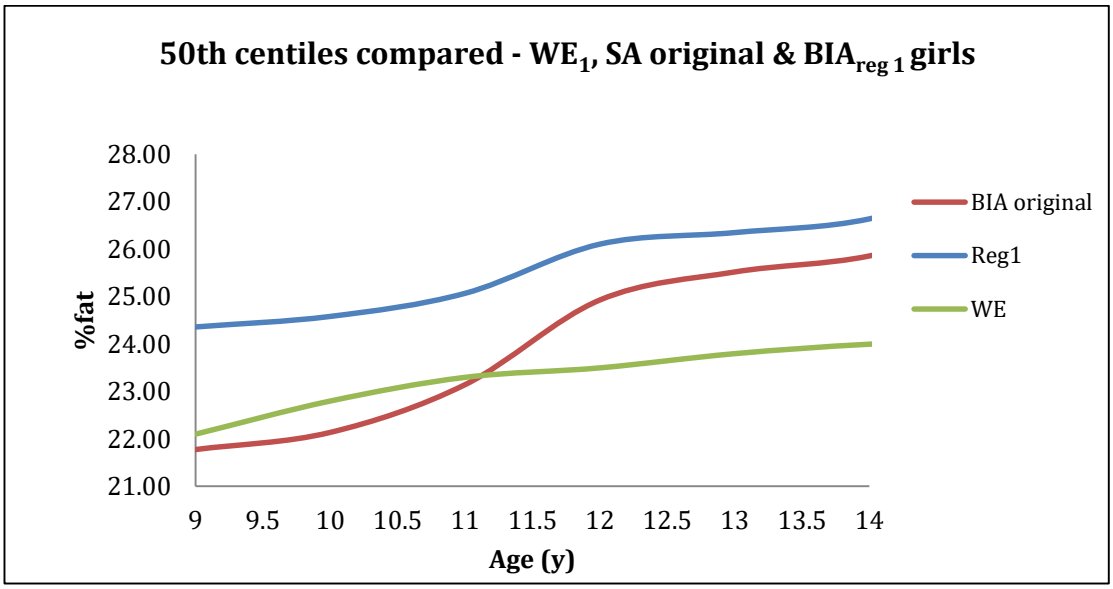


Figure 5.3b %FM comparisons between WE_1 and SA girls before and after application of new BIAreg1 (BIA regression equation see section 4.5.3.5); SA = South Asian; WE_1 = White European data from McCarthy et al. (2006) study. Fat mass = fat mass; BIA = bioelectrical impedance analysis

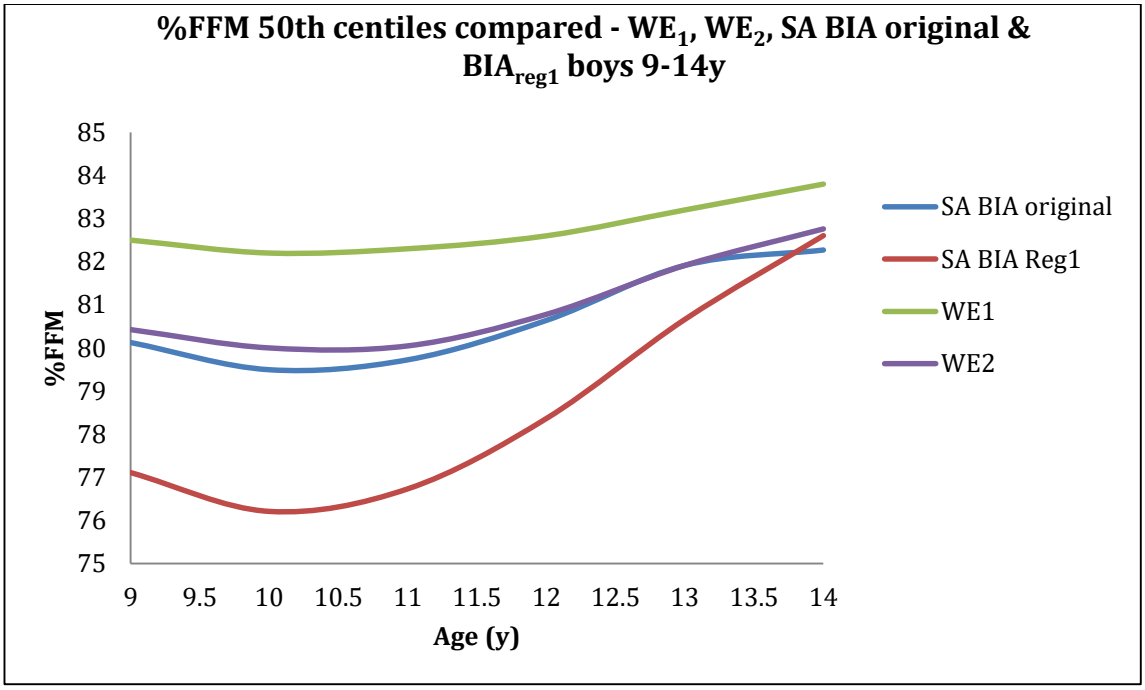


Figure 5.4a FFM (%) centile comparisons between SA, WE_1, & WE_2 boys. FFM = fat-free mass; SA = South Asian; $WE11$= reciprocal of %FM white European data taken from McCarthy et al., 2006; WE2= White European low income data

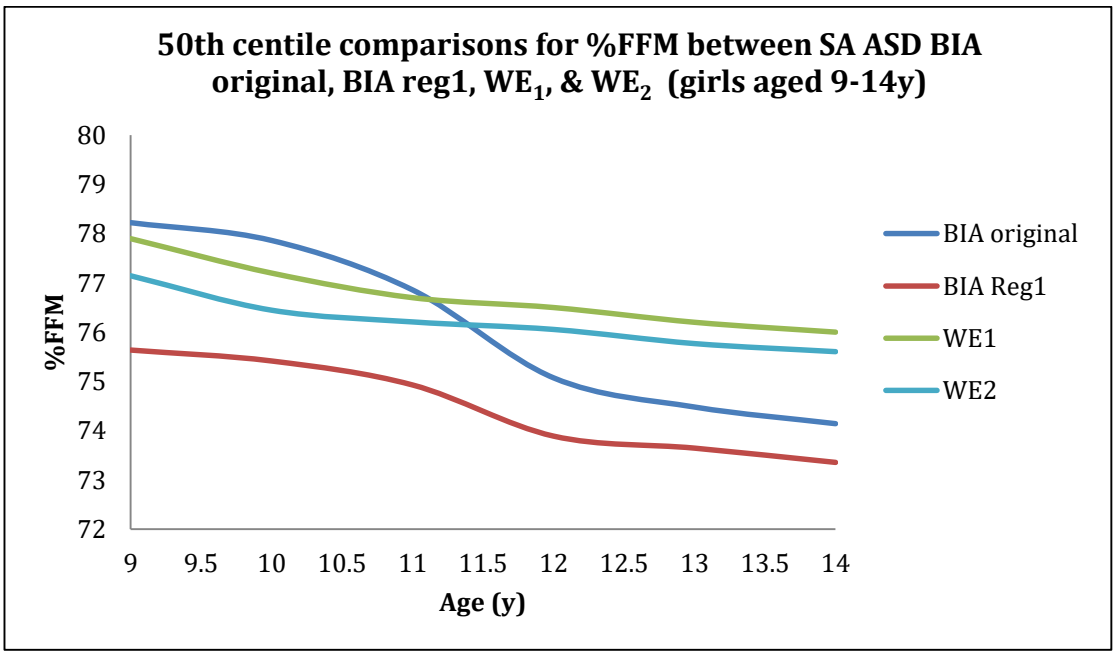


Figure 5.4b FFM (%) centile comparisons between SA, WE_1, & WE_2 girls
FFM = fat-free mass; SA = South Asian; WE_1= reciprocal of %FM white European data taken from McCarthy et al., 2006; WE_2= White European low income data

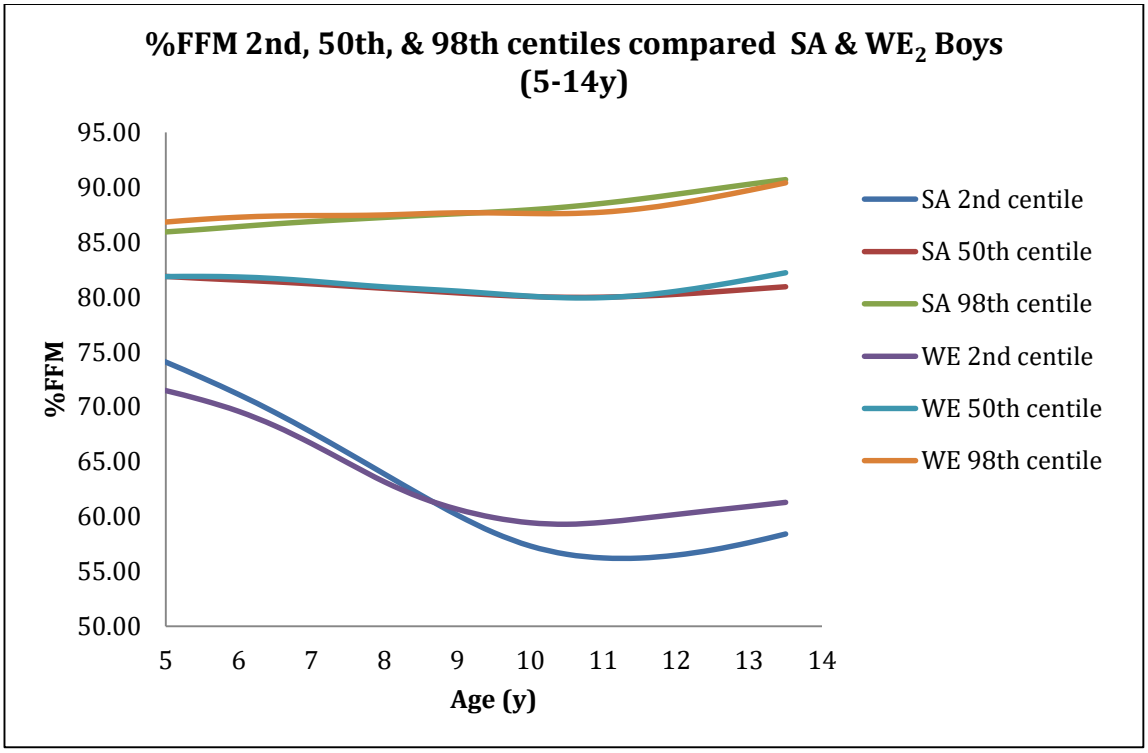


Figure 5.5a FFM (%) centile comparisons between SA & WE₂ boys at the 2nd, 50th, & 98th centiles. WE₂= White European low income data; FFM = fat-free mass; SA = South Asian

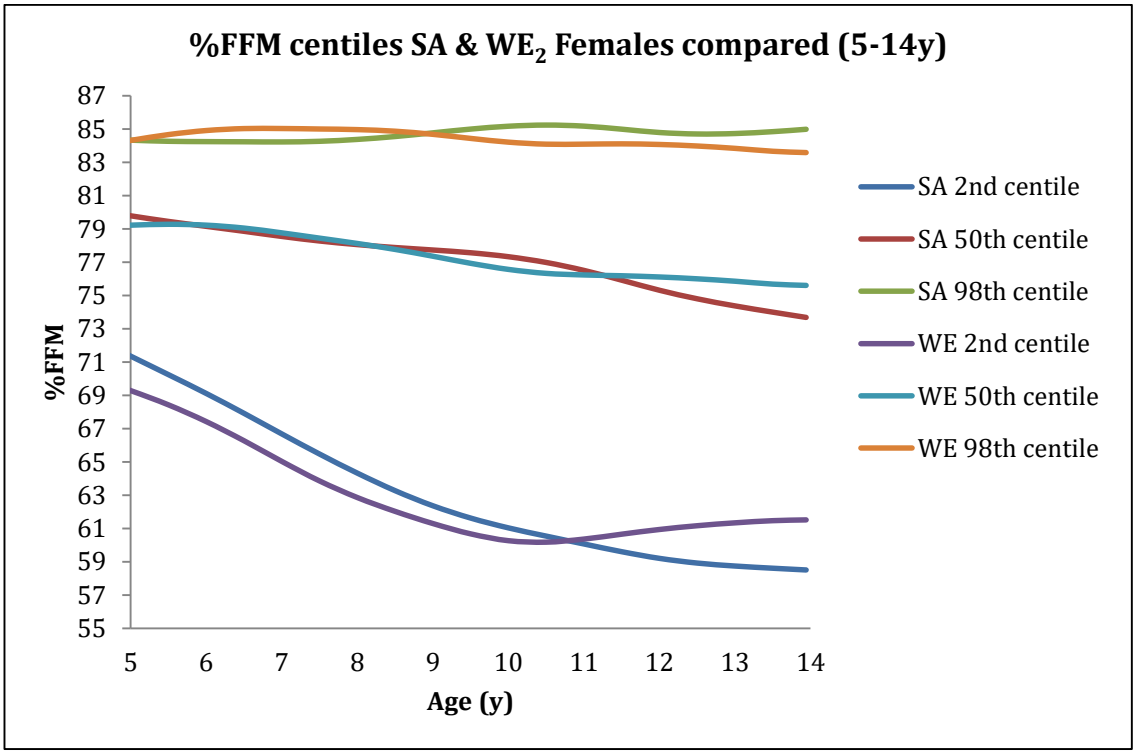


Figure 5.5b FFM (%) centile comparisons between SA & WE₂ girls at the 2nd, 50th, & 98th centiles. WE₂= White European low income data; FFM = fat-free mass; SA = South Asian

No significant differences were observed in %FFM (based on original BIA data; see Table 5.2) between WE_2 and SA boys, whilst for girls, significant differences were only observed in the 11-13y age range, which is evident in the centile graphs in Figures 5.4a and 5.4b for boys and girls respectively. Comparisons at the 50th centile, which included the WE_1 sample (Figure 5.4a) as well as the %FFM for the SA sample following regression (SA BIA_{reg1}), revealed that WE_1 boys had the highest %FFM, with the SA and WE_2 sample, based on the original BIA equation showing almost identical results. Alternatively, as with %FM data (see Figure 5.3a) the SA BIA_{reg1} dataset illustrated how application of the regression equation (to SA children ≥9y) led to a reduced %FFM, with the biggest differences in SA and WE_2 (~ 3%) observed at age 10y, after which the differences appeared to decrease with age. Overall among boys in both SA and WE groups, at the 50th centile %FFM tended to remain relatively constant with age, varying between 80-82% until age 10.5y, and then rose more steeply, rising to ~81% and 82% in SA and WE_2 boys respectively at age 14y, although a steeper incline was observed in the SA BIA_{reg1} dataset.

For girls (Figure 5.4b), use of the original BIA equation resulted in SA girls < age 11y having higher %FFM (the inverse of %FM; Figure 5.3b) than both WE samples, after which it fell below the WE samples. However, the SA BIA_{reg1} dataset indicated that SA girls had less %FFM than both WE groups, with WE_1 having the highest %FFM. The differences between the SA original and BIA_{reg1} output appeared reduced after age 11y. At the 50th centile, %FFM appeared to decline with age in all female groups, with a steeper decline observed between ages 11 -12y in the SA group, compared to both WE groups. Compared to boys in all groups, girls had less %FFM ranging from ~69-79% and 76-79% in SA and WE_2 girls respectively.

Tables showing centiles for measures of %FFM, and measures of SMMa, for SA and WE_2 boys and girls respectively are presented in Appendix E, together with the corresponding reference curves for 7 centiles, and include sex-specific comparisons between WE_2 and SA boys and girls for SMMa measures at the 2nd, 50th, and 98th centiles. SMMa appeared to increase with age for both boys and girls of all groups, although in boys a steeper incline in relative SMMa was observed, whilst in girls it

appeared to plateau at around age 11y although the steepness of the slope was much greater in SMMa/FFM% compared to SMMa%.

5.5 Discussion

This study is the first to our knowledge to generate ethnic specific references for %FM and SMMa, for the SA child and adolescent population in the UK. In general, BIA has been shown to significantly underestimate %FM in the SA population (Haroun at al., 2010; Meyer et al., 2011; Sluyter et al., 2010), and as a result, ethnic-specific prediction equations validated against appropriate reference methods have been recommended. In chapter 4 (BIA Validation Study of Tanita) a prediction equation for determining FFM (kg) using BIA (Tanita BC 418-MA) with DXA as the reference method was successfully developed, from which %FM was determined. However, as the cohort for the validation study had very few children <9y, the equation proved only applicable to children ≥9y; other research has shown that no single equation is valid across the whole child and adolescent age range as the slope and intercept of the regression between Ht^2/Z and FFM changes at different pubertal stages (Montagnese et al., 2013). Therefore, %FM reference curves were only applied to the large external SA dataset of children ≥9y.

It was observed that for SAs, BIA tended to significantly underestimate %FM. In chapter 4, compared to DXA, the underestimation of %FM was 2.23% and 2.79% ($P \leq 0.05$), in boys and girls respectively. In other studies, the degree of underestimation was very similar, with Sluyter et al. (2010) using the same BIA model (Tanita BC 418-MA) as the one used in chapter 4, as well as the same reference method (i.e. DXA), reporting an average underestimation of %FM of 2.84%. A further observation in chapter 4, as well as other studies (Pietrobelli et al., 2004; Sluyter et al., 2010), was that BIA tended to overestimate %FM in leaner subjects and underestimate %FM at high adiposity levels, however, following application of the new prediction equation developed in chapter 4, this discrepancy appeared to be corrected for when comparing the reference curves constructed with new equation to those developed with the original equation, with lower %FM values observed at the 2nd and 9th centiles and higher %FM values observed at the 50th and above centiles, although the degree of difference varied across ages (Tables 5.4-5.7).

Comparisons in %FM reference curves at the 50th centile between SA children from this study and the study on WE_1 (high income) children and adolescents (9-14y) conducted by McCarthy et al. (2006; Figures 5.3a & 5.3b) revealed that for boys, using the original BIA prediction equation, WE_1 children had less %FM overall ranging from a maximum of approximately 3% less fat at age 10.5y to a minimum of approximately 1% less fat at age 14y. Application of the new prediction equation resulted in a very similar shaped curve, however, the differences between SA and WE_1 boys were greater, with WE_1 boys having a maximum of approximately 6% less fat than SA boys at age 10y falling to similar levels as the BIA original equation at age 14y.

For girls, use of the BIA original equation resulted in SA girls aged ≤11y having slightly less %FM (<1%) than WE_1 girls, after which %FM rose much more steeply in SA girls, resulting in SA girls having higher %FM than WE_1 girls with an approximate difference of 2% at age 14y. The outcome in the ≤11y girls was unexpected as the WE_1 dataset used in the McCarthy et al. (2006) study was from a cohort of children from affluent backgrounds, which were considered more likely to have body fat levels similar to the UK90 reference data (i.e. lower %FM levels than the less affluent WEs), to reduce the impact on the outcome of rising levels of obesity; whilst the SA cohort were from more deprived London areas and were thus expected to have higher %FM. Use of the new regression equation however, resulted in SA girls having higher %FM than WE_1 girls across all ages, with an approximate 2% difference at age 9y rising to approximately 3% at age 11.5y. Based on the evidence that BIA underestimates %FM for SAs, and this ethnic group is reported to have higher %FM than WE and other ethnic groups (Ehtisham et al., 2005; Haroun et al., 2010; Nightingale et al., 2011; Rush et al., 2009), suggests that the original BIA equation, particularly for girls in the younger age group (i.e. ≤11y) is unsuitable for SAs.

The %FM centile cut-offs used by McCarthy et al. (2006) to define 'underfat' (2nd centile), 'normal fat' (50th centile), 'overfat' (85th centile), and 'obese' (95th centile) were used to match as closely as possible the IOTF BMI cut-offs (Cole et al., 2000), and similarly were not based on clinical evidence. BMI centile charts lead to higher

levels of children with greater %FM falling within the 'normal' or 'healthy' range. Clinical research would be required to determine appropriate body-fat cut-offs for child and adolescent populations, which may vary by ethnicity due to BC and fat-patterning differences.

Further SA and WE_1 comparisons (9-14y) were made between %FFM at the 50th centile using the reciprocal of %FM data from the McCarthy et al. (2006) study and the low income WE_2 dataset used in this study, to ensure that the ethnic differences in %FM were not simply due to differences in SES (Figures 5.4a and 5.4b). Based on the original BIA equation, for boys, WE_2 children had less %FFM (up to a maximum of ~2% at age 10.5y) than the WE_1 dataset and almost identical levels of %FFM to SAs. However, use of the new regression equation revealed that SA boys also had less %FFM than WE_2 boys, with a maximum difference of approximately 4% at age 10y, with very similar levels of %FFM at age 14y. For girls, the WE_2 dataset also had less %FFM than WE_1 girls, although the differences were very small (~1% maximum), and based on the original BIA equation led to SA girls ≤11y having even greater levels of %FFM, after which age the levels of %FFM fell below WE_2 levels (~1.5% maximum). However, with the new BIA regression equation SA girls of all ages had less %FFM than both the WE_1 and WE_2 datasets with a maximum of approximately 2% less FFM than the WE_2 cohort from 11.5-14y.

Whilst no significant between-group (i.e. SA & WE_2) differences were observed in %FFM (Table 5.2) apart from the SA female group in the 11-13y age range who had significantly ($P<0.05$) less %FFM (1.58%) than their WE_2 counterparts, additional comparisons were made to assess whether application of the BIA_{reg1} equation limited to ages 9-14y would have a significant impact on this result (Table 5.3). Whilst BIA_{reg1} led to a small reduction in the estimation of %FFM for both SA boys (mean difference BIA – BIA_{reg1} = 1.15%; SD = 3.40%) and girls (mean difference BIA – BIA_{reg1} = 0.29%; SD = 9.72%), leading to an increase in between group differences from 0.67% (BIA original) to 1.82% (BIA_{reg1}) among boys and from 0.44% to 0.73% among girls, this difference was only significant ($P=0.009$) between the male groups.

More stable and higher levels of %FFM at the 50th centile among boys (~80-83%) across all ages were observed, compared to girls whose levels of %FFM (~69 -79%) declined with age (Figures 5.4a, 5.4b, 5.5a, & 5.5b), with increasing differences between the sexes observed from age 11y onwards, which are likely attributed to pubertal changes (Wells et al., 2010). Among boys %FM generally decreases more rapidly just before the adolescent growth spurt (~ age 13-15y), reaching its nadir at age 16-17y, with a corresponding increase in %FFM, with gains in muscle mass and bone mineral (Malina, 2005). Among girls, the decline in %FFM with a corresponding increase in %FM is associated with the requirement of additional body fat to support normal reproductive function (Malina, 2005).

Unlike the outcome for %FFM (based on original BIA data), where no significant between-group differences were found apart from in the female 11-13y age range, both SA boys and girls had significantly less relative SMMa (i.e. SMMa%; SMMa/FFM%; SMMaI). For MFR (i.e. SMMa (kg)/FM (kg), significant differences were observed in boys in the 5-7y age range only, and in girls in the 5-7y and 11-13y age range, although across all age ranges both SA boys and girls had lower levels of MFR than their WE_2 counterparts, which has also been observed in other studies (Lear et al., 2009; Rush et al., 2009). Adiposity alone is not the main contributor to T2DM or insulin resistance, particularly among SA children within the normal BMI and Wt ranges (Lear et al., 2009; Whincup et al., 2002; Whincup et al., 2010), with muscle tissue being far more active in glucose uptake, with greater sensitivity to insulin than adipose tissue (Kim et al., 2011; McCarthy et al., 2014).

To our knowledge, this is the first study to make SMMa comparisons between SA and WE populations. The significant findings of ethnic differences in relative SMMa helps to further our understanding and adds to the body of evidence that SA children and adolescents may be at increased risk of obesity related diseases independent of %FM and at lower BMI cut-offs (Saxena et al., 2004; Viner et al., 2009; Whincup et al., 2002).

Significant within-group differences were also observed between Indian, Pakistani, and Bangladeshi children, together with different sub-group outcomes when

compared to the WE_2 cohort for several measures apart from %FM (SDS), which revealed no significant between-group differences (Tables 5.4 & 5.5). For boys, Indian boys had the greatest significant between-group differences, particularly in the 5-7y and 8-10y age ranges, than either Pakistani or Bangladeshi boys for most variables including SDS measures for Ht, Wt, BMI, WC, and absolute measures of SMMa, with lower outcomes in general. For MFR, Indians had the lowest within-group ratio in the 5-7y and 11-13y age ranges, but had the highest MFR in the 8-10y age range together with highest levels of %FFM. Compared to WE_2 boys, Bangladeshi boys were significantly shorter, lighter, with less SMMa (SMMa% and SMMa/FFM%) in one or more age ranges, with no significant differences in BMI. Pakistani boys however had the least differences compared to the WE_2 boys for most measures, although they had the highest sub-group values for BMI, WC and %FM SDS in the 5-7y and 8-10y age ranges, but significantly less SMMa (%) than WE_2 boys in these age ranges.

The sub-group outcomes observed among the girls were similar to the boys, where Indian girls across one or more age ranges were significantly shorter, lighter, with a lower BMI, and less SMMa (both absolute and relative) than WE_2 girls. Indian girls also had less relative SMMa (SMMa/FFM% and SMMaI) than Bangladeshi girls in the 5-7y and 8-10y age ranges. Pakistani girls were also the sub-group with the least differences when compared to the WE_2 girls, although they also had significantly less relative SMMa across two age ranges (5-7y & 11-13y), and were significantly lighter in the 5-7y age range. Similar to the comparisons between SA and WE_2 groups as a whole, all three SA sub-groups in either sex, when compared to their WE_2 counterparts, had no significant differences in %FM, and BMI was either lower or not significantly different, whereas, all sub-groups had significantly less SMMa in one or more age ranges for one or more relative measures. For MFR, whilst no significant between group differences were observed, Pakistani girls had the highest MFR in the 5-7y and 8-10y age ranges, but had the lowest MFR in the 11-13y age range, although the SMMaI was the highest.

In this study, within- and between- sub-group variations in anthropometric and BC measures at different age ranges made it difficult to draw any conclusions on overall

relative health risks by sub-group, although as reported above Pakistani boys in the 8-10y age range had the highest SDS for BMI, WC, and %FM, with significantly less %SMMa than their WE_2 counterparts, which would indicate an increased risk of metabolic syndrome for this sub-group. Clearly there are differences in BC within the SA sub-group, as T2DM is reported to be more prevalent in Pakistani and Bangladeshi ethnic sub-groups in the UK (Bhopal et al., 1999; Erens et al., 2001; Sproston & Mindell, 2006; Wild et al., 2007), with other studies reporting that Bangladeshi children have a more adverse health profile in relation to T2DM than Indian or Pakistani children (Whincup et al., 2010). These differences between studies suggest that variations in BC within group are more likely to be associated with lifestyle factors, such as diet, physical activity, and socioeconomic status (SES), as has been reported in other research (Bhopal et al., 1999; Magnhild et al., 2008; Murasko, 2009; Owen et al., 2009; Saxena et al., 2004). Although, it is also important to note that SAs are a very diverse ethnic group, and such sub-divisions may be too simplistic (Bhopal & Donaldson, 1998). Whilst BC sub-group comparisons led to inconsistent outcomes across different age-ranges, all three sub-groups when compared to their WE_2 counterparts had a lower BMI with no significant differences in %FM, with significantly less relative SMMa in one or more age ranges, and a lower SMMaI across all age ranges for both sexes.

This study adds to the body of evidence highlighting the inadequacies of the use of BMI centiles for identifying overweight or obese children, particularly for the SA population, as the results revealed (Table 5.1) that compared to WE_2 children, SA children in general were significantly ($P<0.001$) shorter, lighter, had a lower BMI and WC (based on SDS data), with no significant differences in %FM (SDS), which indicates that the SA children had greater %FM and thus less %FFM than WE children at the same BMI level. Other studies have also reported similar findings, with SAs having greater %FM and less LM at equivalent BMIs than their WE counterparts (Ehtisham et al., 2005; Haroun et al., 2010; Lear et al., 2009; Nightingale et al., 2011; Rush et al., 2009).

Where SMMa was adjusted for Ht, i.e. SMMaI, this was significantly lower in SAs than the WE_2 group indicating SAs had significantly less appendicular LM, whereas BMI

was also significantly lower in SAs, which as a measure for adiposity would indicate the reverse. Thus SMMaI could be a more useful measure than BMI, similar to the FFMI (Dulloo et al., 2010) or FMI (Whincup et al., 2010) used in other studies, to help identify individuals at risk of T2DM that fall within the normal BMI range.

The current NICE guidance (2013) for determining overweight and obesity among children and adolescents is limited to the use of the BMI centile cut-offs, and it advises the use of caution for the SA population, when interpreting BMI SDS based on the UK90 reference data; whilst WC percentile charts have been recommended by the IDF (Alberti et al., 2007) as an independent predictor of metabolic syndrome. However, based on the findings of this study and other research, neither BMI (Ehtisham et al., 2005; Haroun et al., 2010; Lear et al., 2009; Nightingale et al., 2011; Rush et al., 2009; Whincup et al., 2010) nor WC (Whincup et al., 2010) would provide sufficient information on the obesity related risks within the SA child and adolescent population.

5.5.1 Study Limitations

Due to too few children <9y of age in the BIA validation study (chapter 4), and a lack of data for children ≥14y in the large SA dataset used in this study, the %FM and %FFM reference curves were limited to children aged 9y -14y. The number of children in some of the sub-group comparisons was also very limited, particularly within the Pakistani sub-group, which may have impacted the statistical outcomes. Further studies would be required with a larger dataset for children in the 14-18y-age range, with more equal numbers within each sub-group across all age ranges. Additionally, a valid prediction equation would need to be developed for SA children <9y of age, in order for %FM reference curves to be constructed across the whole child and adolescent age range. %FM and anthropometric (including BMI) comparisons between WE and SA children were based on SDS from the UK90 reference data, however, as there was no similar reference data available for %FFM or SMMa measures, comparisons were made by age-range which is not as accurate.

Whilst both the SA and WE_2 participants in this study were mostly from deprived inner-city London areas, SES was not fully assessed. Besides ethnicity and SES,

several other factors may influence BC, for example, pre-natal and post-natal factors, diet (including infant feeding practices), culture, religion, and physical activity behaviours (Bhopal et al., 1999; Magnhild et al., 2008; Murasko, 2009; Owen et al., 2009; Saxena et al., 2004). Further studies would be required to make a more detailed assessment on the BC differences between SA (as well as within group differences) and WE children and adolescents when all possible confounding variables are properly controlled for, so that any future health recommendations are based on modifiable risk factors.

5.5.2 Conclusion

This study provides valuable BC information for SA children up to age 14y, which would be useful in clinical practice and public health surveillance, together with important comparisons with WE children, particularly how SA children have less relative SMMa and greater %FM at equivalent BMI levels. This research adds to the body of evidence that the current use of BMI is unsuitable as a measure of adiposity among the SA child and adolescent population, and additional measures of WC will not be sufficient to identify individuals at risk of developing metabolic syndrome.

CHAPTER 6: Developing waist circumference percentile charts for South Asian children & youths designed to pass through adult cut-offs

6.1 Introduction

Waist circumference (WC) is a simple and accurate measure of abdominal fatness (Bosy-Westphal et al., 2006; Daniels et al., 2000; Pandey et al., 2009; Shen et al., 2006, Taylor et al., 2000), with evidence that in the UK over the last 20 years, WC has increased more steeply than body mass index (BMI; weight (kg)/height (m^2)) among children and adolescents (McCarthy et al., 2003; Schwandt et al., 2010). Whilst BMI is commonly used as a measure for determining overweight and obesity worldwide (NHS, 2011; WHO, 2000), it is well recognised that it cannot differentiate between individuals with high muscle mass or excess body fat relative to weight, nor does it provide an indication of the distribution of body fat; thus for adults, measurement of WC is also recommended in addition to BMI (NICE, 2014; WHO, 2011). Even among children compared to BMI, WC has been more strongly and independently linked to components of the metabolic syndrome (McCarthy, 2014; Schwandt et al., 2010) such as an adverse atherogenic lipoprotein profile (Flodmark et al., 1994), a raised fasting insulin concentration (Freedman et al., 1999), elevated blood pressure (Choy et al., 2011), and non-alcoholic fatty liver disease (Manco et al., 2008), however, in the UK use of WC cut-offs are currently not recommended (NICE, 2014).

There is much evidence that South Asians (SAs) are at considerably greater risk of morbidity and early mortality from type 2 diabetes mellitus (T2DM) and cardiovascular disease (CVD) than their white European (WE) counterparts (Bhardwaj et al., 2008; Bhopal et al., 1999; Bodicoat et al., 2014; Chaturvedi, 2003; Whincup et al., 2002; WHO, 2004). This increased risk has been attributed to SAs having a genetic propensity to abdominal obesity, with increased visceral and subcutaneous fat (Deurenberg et al., 2002; Lear et al., 2009; Luke, 2009; Saxena et al., 2004; Viner et al., 2010), and less fat-free mass (FFM; Lear et al., 2009) than WE populations, at equivalent BMIs. These ethnic differences in body composition have also been identified in children and adolescents (Ehtisham et al., 2005; Nightingale et al., 2011), with reports in the UK that SA children <16y are almost 14 times at

greater risk of developing symptoms of metabolic syndrome than their WE peers, reflecting SA adult prevalence rates of T2DM (Whincup et al., 2010).

In a UK-based study conducted by Ehtisham et al. (2005), on ethnic differences in body composition and insulin resistance, WC and BMI of SA and WE adolescents aged between 14-17y was measured. Despite a small sample size, compared to the WE cohort (n=31 males and 33 females), SA adolescents in both sexes (n=32 males and 33 females) had significantly ($P<0.05$) higher mean WC measures (79.9cm ±SE2.8 vs 73.4cm ±SE1.7) for males and (71.9cm ±SE2.1 vs 68.1cm ±SE1.5) females respectively. The prevalence (%) of overweight and obese (determined by BMI based on IOTF guidelines) among the SA cohort was also significantly ($P< 0.01$) higher than the WE cohort for both males (41% vs 23%) and females (42% vs 12%).

Wardle et al. (2006) conducted a 5-year longitudinal study measuring the adiposity (WC and BMI) of 5863 ethnically diverse (including Asian n= 175 girls and 253 boys and WE n= 1010 girls and 1597 boys) children from London schools aged 11y at baseline and tracked through to age 16y. In that study WC increased on average by 2.31cm/y (SE 0.09), with the increase in boys' WC being 0.992cm (SE 0.082) greater than girls. Asian girls had the smallest WC ($P<0.001$), with no significant differences between the boys. No significant differences were observed in the prevalence of overweight or obesity (determined by BMI) between Asian and WE children of both sexes when averaged over the 5 years, with 21.3% and 29.4% of boys and girls respectively overweight and obese.

The high prevalence of TDM and CVD, associated with the obesity epidemic, particularly among SA adults (WHO, 2011), and the increasing prevalence of metabolic syndrome and overt T2DM among SA children (Ehtisham et al., 2000; see literature review section 2.2), demonstrates the importance of developing early detection and intervention strategies to prevent or at least reduce the prevalence of these diseases in future populations, particularly among SAs identified as a high-risk population group (IDF, 2007; Misra et al., 2007; Murasko, 2009; Saxena et al., 2004).

To address the high-risk of cardiometabolic diseases among SA adults lower BMI thresholds for overweight and obesity have been recommended by WHO (2004) (see Literature Review section 2.3.1), however, due to a lack of data in ethnic groups other than WE populations, ethnic-specific WC cut-offs were not recommended (WHO, 2011); which is the position adopted by NICE (2014). However, the IDF (Alberti et al., 2006), recommend lower WC cut-offs for SA men compared to WE men (≥90cm & ≥94cm respectively) for diagnosing metabolic syndrome, with a single WC cut-off (≥80cm), for women of all ethnic groups. In 2009, India adopted new BMI cut-offs and two separate WC cut-offs for men and women related to risk of morbidity associated with obesity (chapter 2, section 2.3.1) for its adult population (Misra et al., 2009). A WC cut-off of >78cm for men and >72cm for women was proposed at 'Action level 1', where individuals above this cut-off should be advised to maintain physical activity and avoid gaining weight. Secondly, a WC cut-off of >90cm for men and >80cm for women was proposed at 'Action level 2', where individuals above this cut-off should be advised to seek medical help for further investigation and management of obesity related morbidity.

In the UK an expert group (Viner et al., 2010) voted not to lower BMI thresholds for SA children and adolescents in line with the WHO (2004) 'public health actions points', even though this would lead to some SA adolescents on turning 18y being classified from normal to obese overnight based on the lower SA adult BMI cut-offs (see also Chapter 2 section 2.2.1 and 2.3.2). However, at that time the group recommended that further research was necessary on ethnic differences in body composition (BC) and the metabolic risks associated with adiposity.

Overweight and obesity among children and adolescents is defined by sex-specific BMI centile curves, to account for the changes in growth patterns during this period as well as between genders (Cole et al., 1995; NHS Information Centre, 2011). Currently NICE (2014) do not recommend WC measures to define overweight or obesity among children in the UK. The IDF (Alberti et al., 2007), however, consider WC to be a major predictor of metabolic syndrome for both adults and children, and have recommended use of ≥90th percentile WC cut-off (or adult cut-off if lower in children aged 10-<16y) for determining central obesity in children aged 6-<10y, and

for diagnosing metabolic syndrome in children and adolescence aged 10 <16y. In addition to WC cut-offs, the IDF consensus definition for diagnosing metabolic syndrome requires two or more clinical measures (e.g. elevated triglycerides and raised plasma glucose) for children aged 10y or above. The IDF (Alberti et al., 2007) suggested that children under 10y with abdominal obesity, should not be diagnosed with metabolic syndrome, but be strongly advised on weight reduction.

Several studies have produced gender- and ethnic-specific WC percentile charts of nationally representative samples from across the world including the USA (Fernandez et al., 2004), Italy (Zanolli & Morgese, 1996) and Spain (Moreno et al., 1999) among others. In 2001 McCarthy et al. developed the first British WC percentile curves for WE children and adolescents. Further studies have been recommended to evaluate the health risks associated with abdominal obesity among children and adolescence of different ethnicities, due to evidence of variations in growth and distribution of adiposity through early infancy, childhood and adolescence (Ehtisham et al., 2005; Alberti et al., 2007; McCarthy, 2006; McCarthy et al., 2001). There is clear evidence to support the development of SA child and adolescent WC centile charts similar to those developed for the UK WE population, given that SAs have been identified as a high-risk population for CVD and metabolic syndrome independent of BMI, and in view of the fact that WC thresholds have been revised downwards for SA adults.

6.1.1 Study Aims

The aims of this study were:

i) To develop sex-specific SA WC percentile curves for children and adolescents (5-18y).

ii) To assess within-group differences for WC between Indian, Pakistani, and Bangladeshi children and adolescents.

iii) To compare the results of this study to WE WC percentile curves developed by McCarthy et al. (2001).

6.2 Methods

6.2.1 Participants and Recruitment Procedures

Children and adolescents recruited for the bioelectrical impedance analysis (BIA) validation study for body composition (chapter 4) were also requested to participate in measurements for WC, which was included in the participant consent forms. Further WC measures were taken of children and adolescents as part of the field study, which included BIA and SH measures (chapters 5 & 8 respectively). A total of 126 healthy children and adolescents aged 5-22y were directly measured (86 males and 40 females). Participants were requested to specify ethnicity on their consent forms, and those that had parents from more than one SA ethnic group (e.g. Indian and Pakistani) were defined as mixed SAs. All measures were taken following participant consent as explained in the recruitment procedures in chapter 3 (General Methods). To add to the sample size, data was also taken from a large existing data set of 1459 SA school children and adolescents from inner city London boroughs (Hackney, Tower Hamlets, and Newham) collected at this University for a previous study between 2004 and 2007.

For comparison with WE data, a further existing dataset of 1120 WE school children recruited from schools from the same inner city London Boroughs, which formed part of the previous study as stated above, was also uploaded. The SAs were predominantly second-generation children, and both the WE and SA cohort were from areas of high social and economic deprivation; which limited the possibility of differences in socioeconomic status influencing the between-group anthropometric outcomes.

Due to a lack of data for SA children aged 14y and above, data from the HSE (2004) was used to supplement the data set. However, the sample size was extremely small to influence the overall data outcome.

6.2.2 Anthropometric methods

WC was measured to the nearest 0.1cm using a flexible non-elastic tape measure. The author of this study took all WC measurements, over a single layer of clothing

approximately midway between the top of the iliac crest and lower border of the bottom rib, in accordance with standard procedures (Heyward & Wagner, 2004), and 0.5cm was deducted from the measurement to account for clothing (McCarthy et al., 2003). More detailed methodology is provided in chapter 3 for all measurements taken (General Methods). Measurements for the large data set of WE and SA children and adolescents, that were taken from a previous study at this University, were also carried out by a single researcher, following the same measurement procedures as in this study, to limit inter-observer error.

Sex-specific WC percentiles were constructed using the LMS method as explained in Chapter 3 (General Methods section 3.4.2), based on UK90 growth reference data, and 'Action levels 1 and 2' (i.e. >78cm and 90cm for men; and >72cm and >80cm for women respectively), were identified as WC cut-offs closest to the Indian adult cut-offs at age 16y (Misra et al., 2009). The 90th percentile was also determined as recommended by the IDF (Alberti et al., 2007) and compared with the WE data.

Wt and Ht were measured as described in General Methods (chapter 3), from which BMI (kg/m^2) was calculated. For BMI data, children were classified as overweight or obese at the 85th (SDS 1.04) and 95th (SDS 1.64) percentile cut-offs respectively, as recommended in the National Obesity Observatory (NOO) guidance (NHS, 2011) for population monitoring using the UK90 (Cole et al., 1995) BMI reference data.

6.2.3 Statistical methods

Data were analysed using Microsoft Excel 2011 and SPSS software version 22. Data are presented as means ±SD. Sex-specific between group (SA and WE) and sex- and ethnic-specific (i.e. Indian, Pakistani, Bangladeshi, and mixed SA) within- and between-group (i.e. specified SA ethnicity and WE) comparisons for all anthropometric measures presented in SDS format were conducted on the whole sample using independent samples t-tests and one-way ANOVA respectively.

6.3 Results

Of the cohort that was directly measured (n=126), 6 subjects were aged above 16.99y (4 boys and 2 girls). The additional data from the 1459 (584 boys and 875 girls) SA school children aged 4-16y, had missing WC data for 4 children (2 boys and

2 girls). For the whole dataset combined (n=1581; 913 girls and 668 boys), the majority of children fell in the 4-13.99y age range, therefore, due to a lack of data for children aged 14y and above (for boys 14-14.99; 15-15.99; 16-16.99 n= 7, 6, and 3 respectively; and for girls 14-14.99; 15-15.99; 16-16.99 n= 3, 5, and 0 respectively), data from the HSE (2004) was added to the data set as stated in the methods section above. However, this dataset also had very limited WC data for this age range (for boys 14-14.99; 15-15.99; 16-16.99 n= 1, 2, and 0 respectively; and for girls 14-14.99; 15-15.99; 16-16.99 n= 2, 2, and 2 respectively). As the IDF (2007) recommend using adult cut-offs for adolescents over 16y, and due to a very limited number of individuals aged 17y and over, 16.99y was used as the cut-off age, excluding adolescents above this age from the analysis. The final data set comprised 1593 participants aged 4-16.99y (671 boys and 922 girls).

6.3.1 Characteristics of Study Population

Descriptive characteristics of the cohort (both absolute and SDS) for Wt, Ht, BMI, and WC within each age range are provided in Tables 6.1 and 6.2 for boys and girls respectively, with a breakdown of sample size within the SA ethnicity also shown. For both sexes a very limited number of participants were in the 14y+ age ranges. Limited conclusions could be drawn from such small sample sizes in these latter age ranges, however, in subsequent analyses on the whole dataset (i.e. t-tests, ANOVA), it was considered unlikely to have an impact on the outcome. There were also a very limited number of participants in the 4-4.99y age range, with the majority of Indian descent for both genders. Mixed SAs were the smallest group overall with 54 boys and 76 girls in total.

Up to the age of 13.99y, for boys there was an overall increase in mean Wt, Ht, and WC. BMI however decreased on average from age 4y to 5.99y, and then steadily increased to age 9.99y and appeared to plateau slightly from age 10y to 13.99y, reflecting the typical age-related pattern of BMI. For girls, up to the age of 12.99y all measured variables increased, however, at age 13-13.99y there was a small decline in mean Wt (-1.06kg), BMI (-1.02kg/m^2), and WC (-2.51cm), with only Ht increasing (0.03m).

Table 6.1 Absolute and SDS (mean ±sd) for Wt, Ht, BMI, and WC data of SA boys aged 4.00-16.99y by ethnic sub-group

Age (y)	Sample size (n)	Sample size by ethnicity* (n)	Wt (kg)	Wt SDS	Ht (m)	Ht SDS	BMI (kg/m^2)	BMI SDS	WC (cm)	WC SDS
4.00-4.99	22	1= 16 2= 0 3= 2 4= 4	17.8 (0.19)	-0.39 (1.48)	1.07 (0.05)	-0.11 (1.08)	15.29 (2.35)	-0.50 (1.46)	50.32 (5.48)	-0.58 (1.44)
5.00-5.99	74	1= 42 2= 11 3= 17 4= 4	18.7 (2.53)	-0.52 (1.11)	1.12 (0.04)	-0.15 (0.89)	14.87 (1.38)	-0.68 (1.17)	51.70 (3.93)	-0.20 (1.12)
6.00-6.99	89	1= 34 2= 15 3= 34 4= 6	21.6 (4.13)	-0.37 (1.38)	1.19 (0.05)	-0.05 (1.01)	15.17 (2.05)	-0.54 (1.51)	53.40 (4.99)	-0.02 (1.27)
7.00-7.99	82	1= 31 2= 11 3= 38 4= 2	24.5 (5.18)	-0.18 (1.36)	1.24 (0.05)	-0.10 (0.94)	15.80 (2.57)	-0.21 (1.46)	55.63 (5.99)	0.23 (1.31)
8.00-8.99	69	1= 26 2= 8 3= 30 4= 5	27.9 (6.94)	-0.16 (1.59)	1.29 (0.07)	-0.13 (1.18)	16.37 (2.82)	-0.08 (1.58)	58.42 (7.38)	0.42 (1.42)
9.00-9.99	67	1= 27 2= 10 3= 26 4= 4	31.4 (9.03)	-0.03 (1.33)	1.34 (0.06)	-0.19 (1.05)	17.17 (3.70)	0.09 (1.51)	61.38 (9.45)	0.56 (1.33)
10.00-10.99	81	1= 26 2= 18 3= 32 4= 5	37.8 (11.33)	0.36 (1.46)	1.41 (0.06)	0.09 (0.93)	18.69 (4.42)	0.42 (1.71)	66.25 (11.47)	0.83 (1.50)
11.00-11.99	65	1= 17 2= 20 3= 25 4= 3	39.2 (9.39)	0.15 (1.22)	1.45 (0.06)	-0.10 (0.86)	18.49 (3.75)	0.19 (1.57)	66.86 (10.23)	0.63 (1.33)
12.00-12.99	49	1= 23 2= 9 3= 11 4= 6	45.7 (11.92)	0.32 (1.29)	1.55 (0.09)	0.35 (1.12)	18.80 (3.41)	0.13 (1.36)	67.66 (8.65)	0.48 (1.11)
13.00-13.99	54	1= 27 2= 14 3= 4 4= 9	48.9 (12.14)	0.19 (1.26)	1.60 (0.08)	0.30 (1.00)	18.90 (3.66)	-0.10 (1.47)	67.97 (10.24)	0.15 (1.32)
14.00-14.99	8	1=3 2=1 3=2 4=2	62.4 (13.67)	0.82 (1.02)	1.70 (0.06)	0.26 (0.74)	22.08 (4.55)	0.87 (1.13)	78.78 (9.57)	1.3 (0.86)
15.00-15.99	8	1=2 2=3 3=1 4=2	59.1 (12.60)	-0.01 (1.06)	1.66 (0.05)	-0.69 (0.66)	21.36 (3.60)	0.48 (1.07)	79.45 (11.03)	1.04 (1.28)
16.00-16.99**	3	1= 1 4= 2	51.5 (5.61)	-1.30 (1.01)	1.68 (0.05)	-0.84 (0.73)	18.17 (1.19)	-1.00 (0.85)	68.33 (2.64)	-0.57 (0.64)
Total	671									

SDS data is generated from UK90 LMSgrowth reference data. *Ethnic groups 1= Indian; 2= Pakistani; 3= Bangladeshi; 4= mixed SA. ** Age 16-16.99y taken from HSE, 2004 data. SA = South Asian; Wt, Ht, and BMI were only available for one subject. Wt= weight; Ht = height; BMI = body mass index; WC = waist circumference

Table 6.2 Absolute and SDS (mean ±sd) for Wt, Ht, BMI, and WC data of SA girls aged 4.00-16.99y by ethnic sub-group

Age (y)	Sample size (n)	Sample size by ethnicity* (n)	Wt (kg)	Wt SDS	Ht (m)	Ht SDS	BMI (kg/m²)	BMI SDS	WC (cm)	WC SDS
4.00-4.99	27	1= 20 2= 1 3= 2 4= 4	16.1 (3.80)	-1.02 (1.64)	1.05 (0.05)	-0.30 (1.02)	14.36 (2.31)	-1.17 (2.35)	48.20 (4.94)	-0.93 (1.53)
5.00-5.99	66	1= 33 2= 4 3= 22 4= 7	18.0 (2.44)	-0.68 (1.02)	1.11 (0.05)	-0.33 (0.99)	14.61 (1.41)	-0.71 (1.01)	50.34 (3.63)	-0.35 (1.03)
6.00-6.99	69	1= 27 2= 5 3= 28 4= 9	20.8 (4.46)	-0.53 (1.33)	1.17 (0.06)	-0.18 (1.16)	14.98 (2.39)	-0.64 (1.47)	52.56 (5.92)	-0.13 (1.37)
7.00-7.99	87	1= 35 2= 5 3= 36 4= 11	24.2 (5.33)	-0.23 (1.33)	1.24 (0.06)	-0.05 (1.13)	15.71 (2.27)	-0.25 (1.21)	54.68 (5.72)	0.13 (1.23)
8.00-8.99	73	1= 38 2= 3 3= 27 4= 5	27.0 (6.08)	-0.34 (1.30)	1.29 (0.05)	-0.28 (0.96)	16.19 (2.82)	-0.26 (1.32)	56.40 (6.39)	0.14 (1.29)
9.00-9.99	68	1= 25 2= 4 3= 37 4= 2	31.1 (7.29)	-0.17 (1.34)	1.36 (0.08)	0.09 (1.26)	16.57 (2.78)	-0.27 (1.24)	58.68 (7.43)	0.29 (1.27)
10.00-10.99	63	1= 23 2= 8 3= 25 4= 7	33.0 (7.34)	-0.41 (1.20)	1.40 (0.06)	-0.27 (0.95)	16.80 (2.92)	-0.45 (1.38)	59.31 (7.98)	0.06 (1.48)
11.00-11.99	206	1= 57 2= 69 3= 72 4= 8	41.3 (11.78)	0.13 (1.37)	1.48 (0.07)	0.08 (1.01)	18.75 (4.23)	0.02 (1.48)	63.42 (10.15)	0.41 (1.58)
12.00-12.99	190	1= 74 2= 62 3= 38 4= 16	46.6 (10.52)	0.24 (1.25)	1.53 (0.06)	0.02 (0.89)	19.82 (3.98)	0.20 (1.47)	66.13 (9.66)	0.57 (1.55)
13.00-13.99	61	1= 38 2= 12 3= 8 4= 3	45.5 (7.89)	-0.27 (1.01)	1.56 (0.06)	-0.15 (0.85)	18.80 (3.21)	-0.30 (1.30)	63.62 (6.95)	0.03 (1.36)
14.00-14.99	5	1=3 2=0 3=1 4=1	55.6 (14.11)	0.28 (1.51)	1.60 (0.04)	-0.23 (0.59)	21.74 (4.86)	0.45 (1.50)	74.57 (13.68)	1.43 (1.65)
15.00-15.99	5	1=2 2=0 3=0 4=3	46.2 (12.60)	-1.60 (2.75)	1.51 (0.18)	-1.84 (2.96)	19.67 (1.74)	-0.26 (0.75)	65.84 (6.34)	0.06 (1.34)
16.00-16.99**	2	1=2	52.0	-0.56	1.63	-0.14	19.67	-0.4	79.9 (8.63)	2.15 (1.03)
Total	922									

SDS data is generated from UK90 LMSgrowth reference data. *Ethnic groups 1= Indian; 2= Pakistani; 3= Bangladeshi; 4= mixed SA. ** Age 16-16.99y taken from HSE, 2004 data. SA = South Asian; Wt, Ht, and BMI were only available for one subject. Wt= weight; Ht = height; BMI = body mass index; WC = waist circumference

6.3.2 Waist Circumference Percentiles

Selected sex specific WC percentiles are shown in Table 6.3, for each age range from 4y+ to 16y+. For boys and girls respectively, the 71st and 68th percentiles are highlighted as closest to the Indian adult cut-offs at 15y+ (Misra et al., 2009) at 'Action level 1' (>78cm for boys and >72cm for girls). The 90th and 87th percentiles are also highlighted for boy and girls respectively as closest to the Indian adult cut-offs at 15y+ at 'Action level 2' (>90cm for males and >80cm for females) which are also the IDF (2006) cut-offs for central obesity for SA adult males and females respectively (i.e. ≥90cm and ≥80cm).

Figures 6.1 and 6.2 show the smoothed WC percentile curves for boys and girls respectively with 9 centiles equivalent to the format of the published UK90 BMI reference curves (Cole et al., 1995). Figures 6.3 and 6.4 show the smoothed centiles for boys and girls respectively with 7 centiles, starting at the 9th centile and including the centiles closest to the Indian adult cut-offs at 'Action level 1' and 'Action level 2' as stated above.

Table 6.3 Percentiles of waist circumference (WC) by age and sex

			Percentiles								
SEX	**Age**	**n**	**9th**	**25th**	**50th**	**71st***	**75th**	**90th****	**91st**	**98th**	**99.6th**
BOYS	4+	22	45.58	46.96	48.64	**50.30**	50.71	**53.08**	53.34	57.28	62.35
	5+	74	46.41	48.02	49.99	**51.98**	52.47	**55.37**	55.70	60.71	67.63
	6+	89	47.44	49.38	51.79	**54.25**	54.87	**58.55**	58.98	65.64	75.55
	7+	82	48.49	50.80	53.68	**56.66**	57.42	**61.96**	62.50	70.97	84.26
	8+	69	49.83	52.52	55.92	**59.46**	60.37	**65.86**	66.50	76.98	94.11
	9+	67	51.40	54.48	58.40	**62.53**	63.58	**70.06**	70.83	83.46	104.86
	10+	81	53.05	56.50	60.91	**65.59**	66.79	**74.19**	75.07	89.72	115.07
	11+	65	54.61	58.37	63.20	**68.33**	69.64	**77.77**	78.74	94.77	122.20
	12+	49	56.09	60.11	65.27	**70.75**	72.15	**80.80**	81.83	98.75	126.91
	13+	54	57.52	61.77	67.22	**72.98**	74.46	**83.53**	84.60	102.11	130.42
	14+	8	59.09	63.56	69.30	**75.34**	76.88	**86.32**	87.44	105.40	133.46
	15+	8	60.79	65.51	71.53	**77.86**	79.47	**89.27**	90.42	108.75	136.34
	16+	3	62.44	67.40	73.72	**80.32**	82.00	**92.16**	93.35	112.03	139.28
			9th	**25th**	**50th**	**68th***	**75th**	**87th****	**91st**	**98th**	**99.6th**
GIRLS	4+	27	43.09	44.76	46.77	**48.42**	48.75	**51.23**	52.30	56.74	62.07
	5+	66	44.42	46.28	48.53	**50.39**	50.77	**53.60**	54.84	60.05	66.55
	6+	69	46.01	48.12	50.70	**52.85**	53.29	**56.61**	58.09	64.40	72.61
	7+	87	47.44	49.81	52.75	**55.22**	55.72	**59.59**	61.32	68.85	78.98
	8+	73	48.64	51.30	54.61	**57.41**	57.98	**62.38**	64.37	73.05	84.90
	9+	68	49.62	52.60	56.30	**59.43**	60.07	**65.02**	67.24	76.98	90.16
	10+	63	50.60	53.91	58.02	**61.50**	62.21	**67.69**	70.15	80.80	94.90
	11+	206	51.86	55.52	60.06	**63.89**	64.67	**70.65**	73.32	84.69	99.23
	12+	190	53.48	57.49	62.44	**66.59**	67.43	**73.84**	76.67	88.54	103.12
	13+	61	54.81	59.14	64.43	**68.82**	69.71	**76.38**	79.28	91.18	105.09
	14+	5	55.92	60.56	66.18	**70.77**	71.68	**78.52**	81.44	93.09	106.01
	15+	5	57.09	62.09	68.04	**72.83**	73.77	**80.74**	83.66	95.00	106.95
	16+	2	58.31	63.70	70.01	**75.00**	75.98	**83.06**	85.98	97.00	108.10

Age presented as each complete year e.g. 4+ = 4.00 – 4.99y; percentile values are in cm.

* Percentile closest to adult WC cut-off action level 1 for Asian Indians (Misra et al., 2009) at age 15+y (Boys >78cm; girls > 72cm)

** Percentile closest to adult WC cut-off action level 2 for Asian Indians (Misra et al., 2009) at age 15+y (Boys >90cm; girls > 80cm)

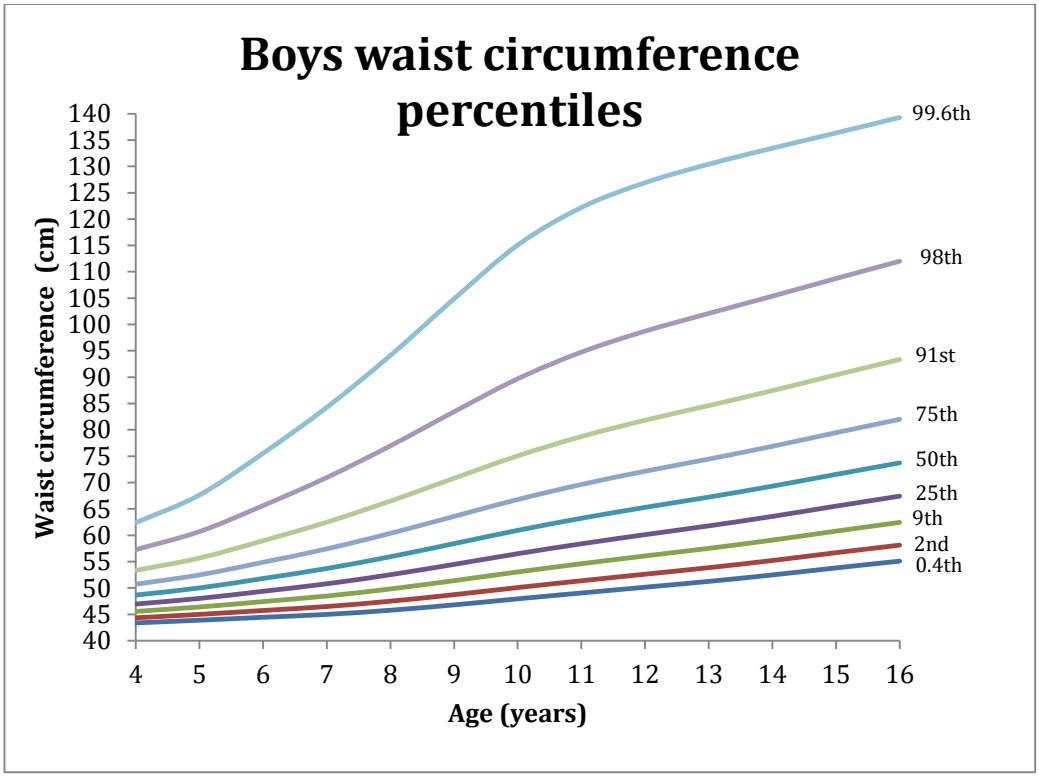


Figure 6.1 Smoothed waist circumference percentile curves with 9 centiles shown equivalent to UK90 BMI centiles for South Asian boys aged 4.00-16.99y

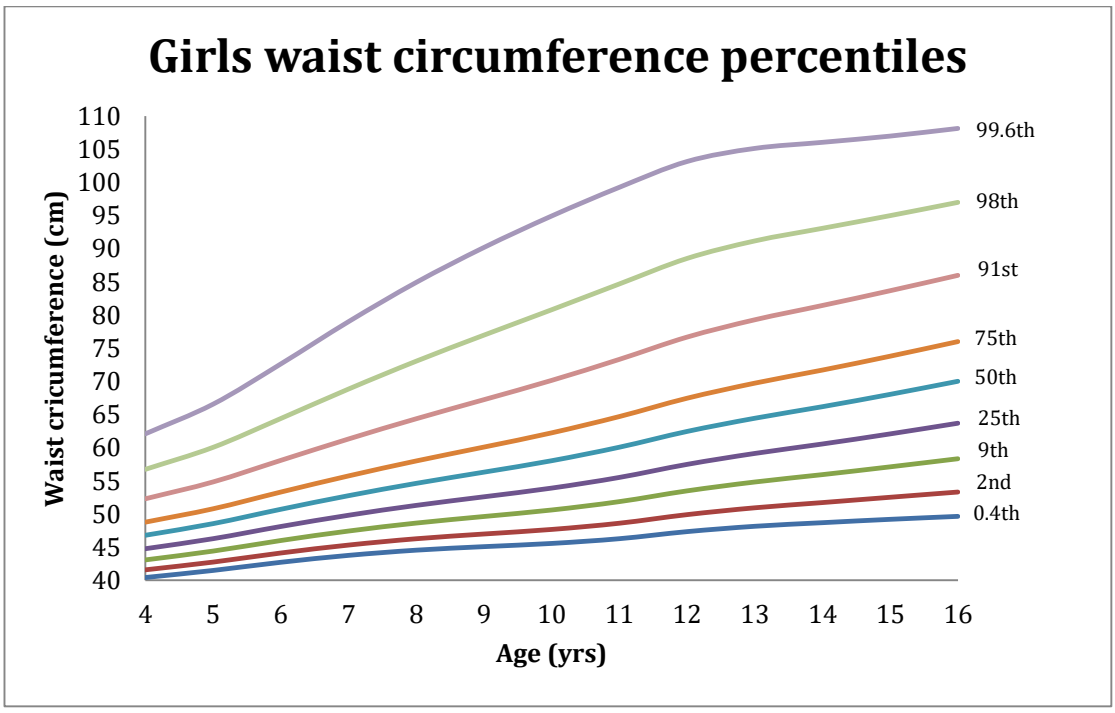


Figure 6.2 Smoothed waist circumference percentile curves with 9 centiles shown equivalent to UK90 BMI centiles for South Asian girls aged 4.00-16.99y

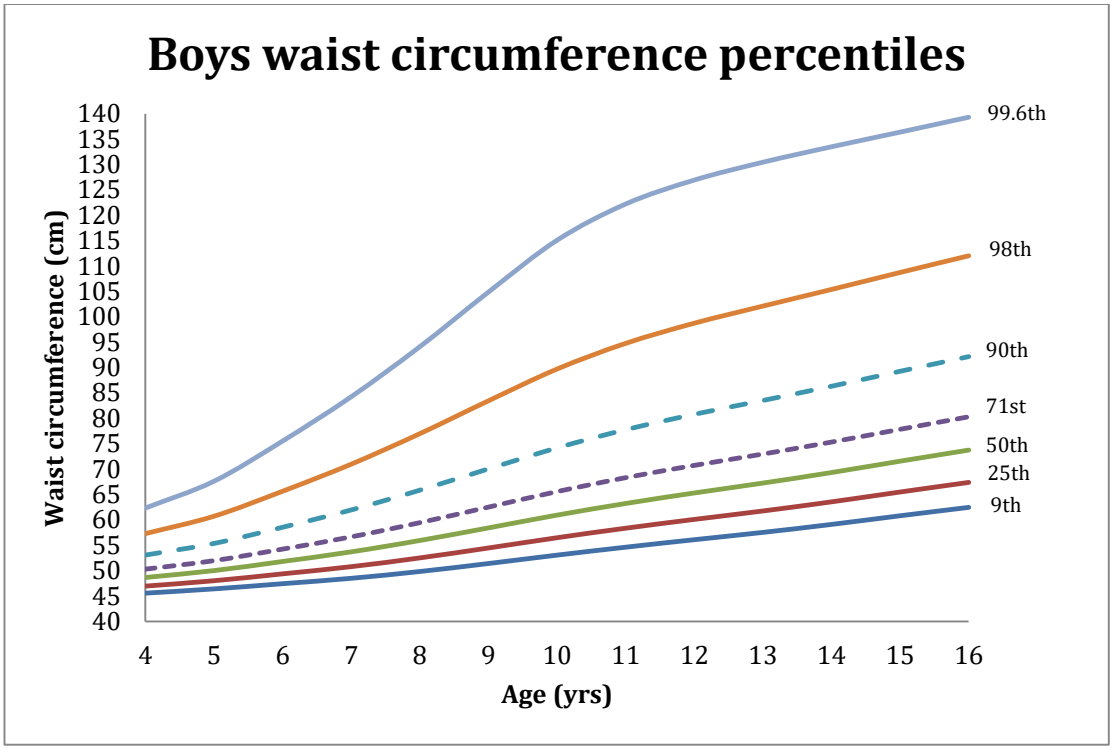


Figure 6.3 Smoothed waist circumference percentile curves with 7 centiles shown – 71st and 90th representing centile lines that pass close to adult cut-offs for 'Action levels 1 and 2' (Misra et al., 2009) respectively for South Asian boys at age 15+y

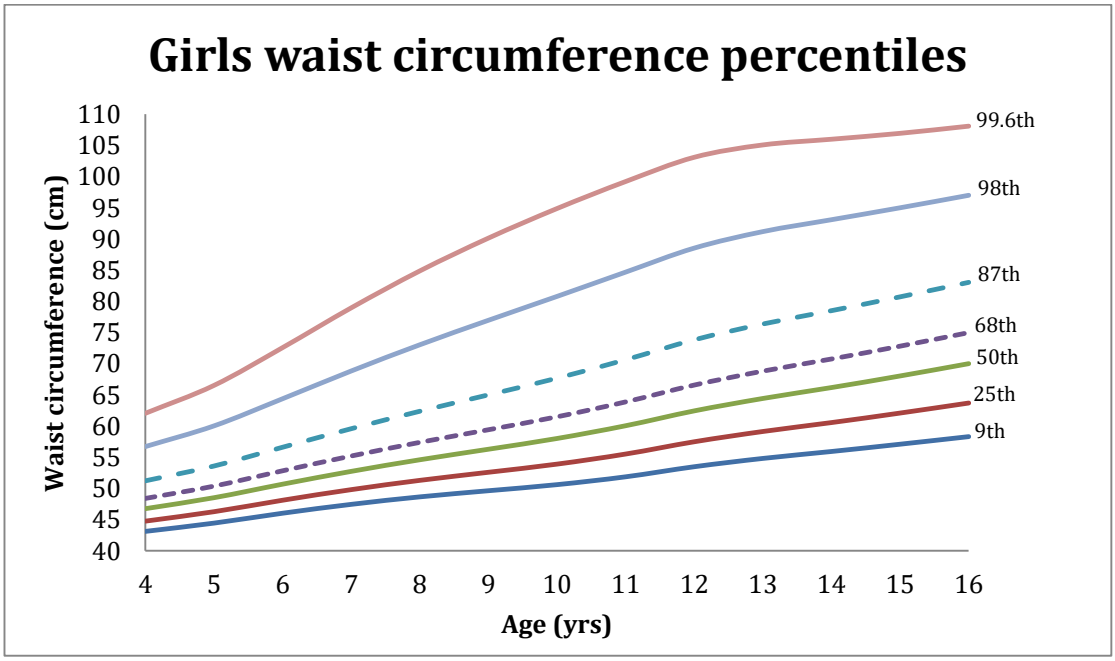


Figure 6.4 Smoothed waist circumference percentile curves with 7 centiles shown – 68th and 87th representing centile lines that pass close to adult cut-offs for 'Action levels 1 and 2' (Misra et al., 2009) respectively for South Asian girls at age 15+y

6.3.3 Waist circumference percentile and anthropometric comparisons between South Asians and white Europeans

Figures 6.5 and 6.6 show comparisons between smoothed centile curves for SA children from this study and UK WE children (McCarthy et al., 2001) at the 50th and 90th centiles from age 5y to 16.99y. Clear differences between SA and WE children were apparent for both sexes, particularly at the 90th centile with SA children having much higher waist circumferences than WE children, with these differences increasing from around age 9y. At the 50th centile both SA children and WE appeared to have similar waist measures up to age 8y. However, for boys the greatest differences appeared between age 9y -13y, after which the differences appeared smaller. For girls the greatest differences appeared at around age 12y and continued through to age 16y+, with the curve showing a slight plateau for the WE girls, whilst for SA girls the centile curve continued to incline upwards, although this may be due to the lack of data for the upper age ranges.

Table 6.4 shows anthropometric comparisons for all SDS (based on UK90 reference data) measured variables between SAs from this study and the large WE dataset of children taken from a previous study at this university. An Independent Samples t-test revealed significant differences between the WE and SA children and adolescents for all the measured variables ($P<0.001$), with SAs being significantly shorter, lighter, with a lower BMI and WC. Compared to the UK90 data, SAs from this study were very similar to the mean (i.e. SDS 0 or 50th centile) for Ht (SDS -0.03), and Wt (SDS -0.05), with a slightly lower BMI (SDS -0.10 or 42nd centile), and higher WC (SDS 0.32 or 63rd centile).

Prevalence of overweight based on BMI (BMI >85th percentile or SDS >1.03) and obesity (BMI >95th percentile or SDS > 1.64) was lower in the SA cohort than the WE cohort for both boys (8.2% vs 10.5% for overweight; and 16.3% vs 18% for obese) and girls (9.9% vs 12.3% for overweight; and 11.2% vs 14% for obese) respectively. A much greater proportion of both SA boys (16%) and girls (15%) were observed at or above the WC cut-off for 'Action level 1' (71st centile for boys and 68th centile for girls), than compared to the BMI overweight cut-offs. Similarly, compared to the BMI obese cut-offs, a much greater proportion of both WE and SA boys and girls were identified at or above the 90th centile (and/or 'Action level 2' for SA boys) for

WC (WE boys = 29% vs SA boys = 25% and WE girls = 27% vs SA girls = 25%). 29% of SA girls were observed at or above the WC 'Action level 2' (or 87th percentile) cut-off. Comparisons of prevalence (%) between the SA and WE samples by z test, of overweight, obese (BMI>85th and 95th percentile respectively), and raised WC (≥90th percentile) revealed no significant differences (at the P<0.05 level).

A further comparison was made to assess the differences in prevalence of BMI and WC at or below the 2nd centile (i.e. SDS <-1.96; Table 6.4). For both SA (SDS <-1.96 = 10.3% vs 2.8% and 9.9% vs 6.1%) and WE (SDS <-1.96 = 3.4% vs 0.5% and 2.3% vs 0.9%) ethnicities a greater proportion of children were observed to be at or below the BMI 2nd centile than compared to the WC 2nd centile for both boys and girls respectively. These differences were significant (P<0.05) for SA and WE boys and SA girls only.

Table 6.5 shows the same anthropometric characteristics for the WE dataset as in Table 6.4; however, for the SA group, data for specified ethnicities within the SA ethnic group are presented, and compared with the WE data using one-way ANOVA. The Pakistani group, compared to their WE peers, was the only ethnicity in which no significant differences were observed for all measured variables for both sexes. However, compared to their WE peers both sexes of Indians and Bangladeshis, and girls only of mixed SA ethnicity had significantly lower SDS for all measured variables (P<0.001 or P<0.05). No significant differences were observed between WE and mixed SA boys for all measured variables apart from WC, with mixed SA boys having a significantly lower (P<0.001) WC than their WE peers.

Compared to the UK90 growth reference data for BMI SDS and WC SDS, all ethnicities within the SA group, including those with lower than average BMI SDS (i.e. <0.00 or 50th percentile) had higher WC SDS than the mean UK90 reference population. Both Indian boys and girls had a mean BMI SDS at the 37th percentile (SDS -0.35), whereas the WC percentile was slightly above the 50th percentile (SDS 0.08 and 0.03 for boys and girls respectively). Bangladeshi boys had a mean BMI close to the 50th percentile (SDS 0.01); however, the mean WC was at the 64th percentile (SDS 0.35), and Bangladeshi girls had a mean BMI at the 43rd centile (SDS-

0.12) and a mean WC at the 60th centile (SDS 0.26). Pakistani boys had a mean BMI at the 59th percentile (SDS 0.22), with a mean WC at the 80th percentile (SDS 0.84), whilst Pakistani girls had a mean BMI at the 52nd centile (SDS 0.06), and a mean WC at the 71st centile (SDS 0.56).

Within group differences were also observed for Ht, Wt, BMI, and WC SDS. For boys: Bangladeshis were significantly shorter than Pakistanis (p=0.002) and mixed SAs (p=0.032); Pakistanis were significantly heavier than Indians (p=0.003); Pakistanis (p=0.003) and Bangladeshis (p= 0.049) had significantly higher BMIs than Indians; for WC Pakistanis had a significantly higher WC than all other SA ethnic groups (Indians P<0.001; Bangladeshis p= 0.007; mixed SA p = 0.019). For girls: for Ht SDS there were no within group differences; for Wt SDS Pakistanis were significantly heavier (p=0.007) than Indians, and mixed SAs (p=0.028); had a significantly higher BMI SDS (p=0.009) than Indians, and a significantly higher (P< 0.001) WC SDS than Indians, and mixed SAs (p= 0.041).

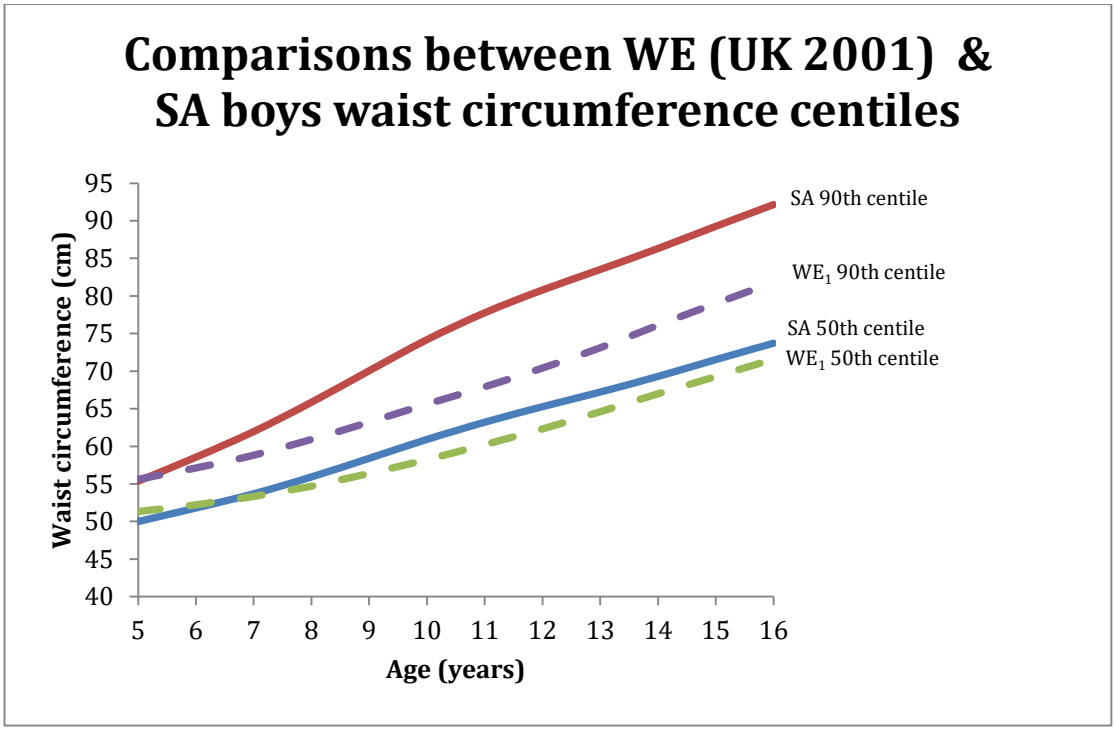


Figure 6.5 Comparisons between UK WE_1 and SA 50th and 90th centiles for boys age 5-16.99y. WE_1 = white European data from McCarthy et al., 2001; SA = South Asian

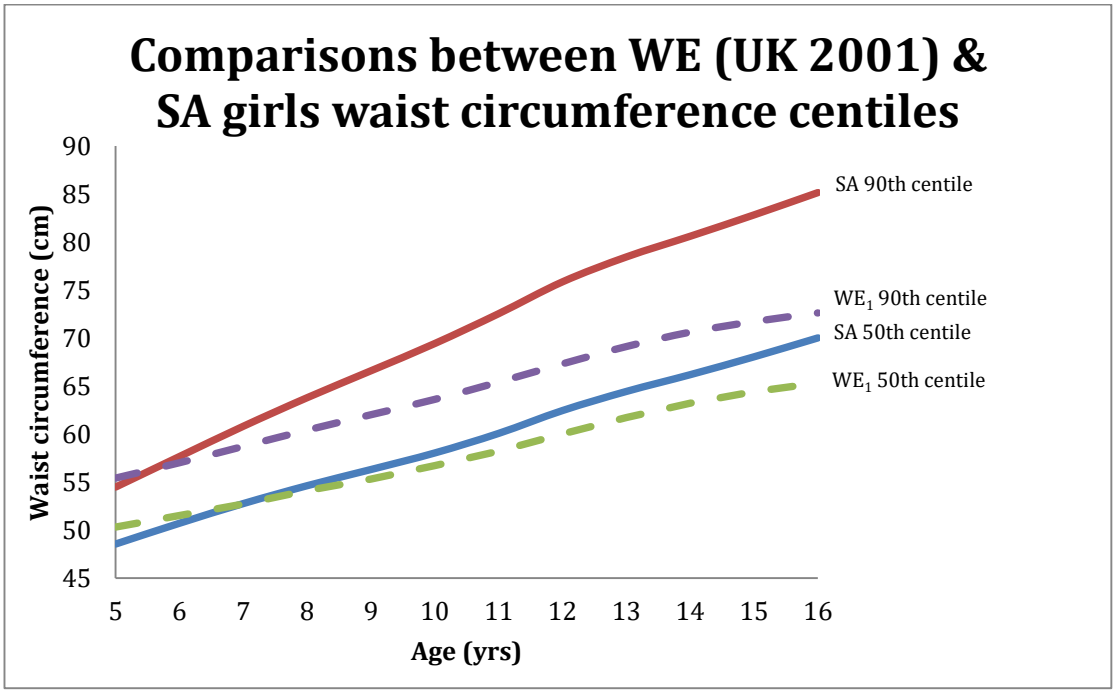


Figure 6.6 Comparisons between UK WE_1 and SA 50th and 90th centiles for girls age 5-16.99y. WE_1 = white European data from McCarthy et al., 2001; SA = South Asian

Table 6.4. Anthropometric characteristics (Ht, Wt, BMI, WC SDS, % with overweight & obese BMI, & % with WC SDS at 2nd centile & at 'Action levels 1$^{\chi}$ & 2$^{\delta}$' (SA only) of SA and WE cohorts by sex

	Boys		**Girls**	
	SA (n= 671)	WE (n=588)	SA (n= 921)	WE (n= 532)
Decimal Age (y)	9.17 (2.77)	8.84 (2.49)	10.05 (2.64)	8.83 (2.43)
Ht SDS	-0.03* (1.00)	0.30 (1.01)	-0.08* (1.04)	0.15 (1.02)
Wt SDS	-0.05* (1.37)	0.44 (1.25)	-0.15* (1.33)	0.22 (1.20)
BMI SDS	-0.10* (1.51)	0.37 (1.34)	-0.20* (1.42)	0.18 (1.26)
BMI Overweight$^{\alpha}$ n (%)	55 (8.2)	63 (10.5)	91 (9.9)	67 (12.3)
BMI obese$^{\beta}$ n (%)	109 (16.3)	108 (18.0)	103 (11.2)	76 (14.0)
BMI SDS <-1.96 n (%)	69 (10.3)	20 (3.4)	91 (9.9)	12 (2.3)
WC SDS <-1.96 n (%)	19 (2.8)	3 (0.5)	56 (6.1)	5 (0.9)
WC SDS	0.32* (1.36)	0.74 (1.17)	0.21* (1.46)	0.64 (1.19)
WC SDS Action level 1$^{\chi}$ n (%)	108 (16)	-	137 (15)	-
WC SDS Action level 2$^{\delta}$ n (%)	168 (25)		269 (29)	-
WC SDS ≥ 1.29 (90th centile)	168 (25)	170 (29)	228 (25)	144 (27)

Data are means (±s.d.) unless otherwise stated. SDS data is generated from UK90 LMSgrowth reference data
Independent Samples Test between SA & WE for both boys and girls significantly different *P<0.001
SA = South Asian; WE = white European; Ht = height; Wt = weight; BMI = body mass index; WC = waist circumference; SDS = standard deviation score; $^{\alpha}$BMI SDS > 1.03 (85th centile) for overweight and $^{\beta}$BMI SDS > 1.64 (95th centile) for obese
$^{\chi}$ WC SDS Action level 1 ≥ 0.56 (71st centile) for boys and ≥ 0.47 (68th centile) for girls; $^{\delta}$ WC SDS Action level 2 ≥ 1.29 (90th centile) for SA boys and ≥ 1.13 (87th centile) for SA girls; for both WE boys and girls the 90th centile cut-off (SDS≥ 1.29) was used and is also shown for SA girls for purposes of comparison with WE girls. WC SDS 'Action levels 1 & 2' based on adult cut-offs adopted by India (Misra et al., 2009).

Table 6.5 Sex-specific anthropometric characteristics (Ht, Wt, BMI, WC SDS, % with overweight & obese BMI, & % with WC SDS at 2nd centile & at 'Action levels 1$^{\chi}$ & 2$^{\delta}$' (South Asian only) of white European and South Asian cohort by ethnic sub-group

	Boys					**Girls**				
Ethnic code	1 (n=274)	2 (n=121)	3 (n=222)	4 (n=54)	5 (n=588)	1 (n=375)	2 (n= 174)	3 (n=296)	4 (n= 76)	5 (n= 532)
Ht SDS	-0.02* (1.07)	0.20 (0.94)	-0.23* (0.97)	0.21 (0.82)	0.30 (1.01)	-0.09** (1.06)	0.03 (1.09)	-1.11** (1.01)	-0.27** (0.96)	0.15 (1.02)
Wt SDS	-0.22* (1.37)	0.30 (1.25)	-0.11* (1.45)	0.18 (1.10)	0.44 (1.25)	-0.27* (1.32)	0.12 (1.40)	-0.12** (1.31)	-0.40** (1.24)	0.22 (1.20)
BMI SDS	-0.35* (1.51)	0.22 (1.46)	0.01* (1.56)	0.02 (1.30)	0.37 (1.34)	-0.35* (1.38)	0.06 (1.55)	-0.12** (1.39)	-0.38** (1.35)	0.18 (1.26)
BMI Overweight$^{\alpha}$ n (%)	23 (8.4)	11 (9.2)	15(6.8)	6 (11.1)	63 (10.5)	31 (8.3)	24 (13.9)	32 (10.8)	4 (5.3)	67 (12.3)
BMI obese$^{\beta}$ n (%)	30 (10.9)	28 (23.5)	43 (19.4)	8 (14.8)	108 (18)	34 (9.1)	28 (16.2)	35 (11.8)	6 (7.9)	76 (14)
WC SDS	0.08* (1.32)	0.84 (1.31)	0.35* (1.37)	0.21* (1.22)	0.74 (1.17)	0.03* (1.44)	0.56 (1.57)	0.26** (1.40)	0.03** (1.33)	0.64 (1.19)
WC SDS Action level 2$^{\delta}$ n (%)	56 (20)	47 (39)	52 (23)	13 (24)	-	96 (26)	73 (42)	85 (29)	15 (20)	-
WC SDS ≥ 1.29 (90th centile) n (%)	56 (20)	47 (39)	52 (23)	13 (24)	170 (29)	78 (21)	64 (37)	74 (25)	12 (16)	144 (27)

SA = South Asian; WE = white European; Ht = height; Wt = weight; BMI = body mass index; WC = waist circumference; SDS = standard deviation score; ethnic codes: 1= Indian; 2= Pakistani; 3= Bangladeshi; 4= Mixed South Asian; 5= White European (WE). Data are means (±s.d.) unless otherwise stated. SDS data is generated from UK90 LMSgrowth reference data
Between-group differences analysed using one-way ANOVA
Mean difference is significant between specified SA ethnic group and WE sample *P<0.001; **P<0.05
$^{\alpha}$BMI SDS >1.03 (85th centile) and $^{\beta}$BMI SDS > 1.64 (95th centile)
$^{\delta}$ Waist circumference SDS Action level 2 ≥ 1.29 (and/or 90th centile) for SA boys and ≥ 1.13 (87th centile) for SA girls; for both WE boys and girls the 90th centile cut-off (SDS≥ 1.29) was used for comparison with SA data. WC SDS 'Action level 2' based on adult cut-offs adopted by India (Misra et al., 2009).

6.4 Discussion

This study, to our knowledge, presents the first WC percentile charts for SA children and adolescents aged 4.00-16.99y. BMI and WC (SDS) comparisons of the SA sample with the UK90 growth reference data (Cole et al., 1995; Table 6.4) revealed that whilst both SA boys and girls had lower than average BMI (SDS -0.10 and -0.20 for boys and girls respectively), WC was higher (SDS 0.32 and 0.21 for boys and girls respectively). This provides further evidence that BMI is limited in identifying populations at risk of morbidities related to overweight rather than over-fat, particularly in SA populations with higher levels of abdominal adiposity linked to an increased risk of metabolic syndrome and T2DM (Deurenberg et al., 2002; Lear et al., 2009; Luke, 2009; Saxena et al., 2004; Viner et al., 2010).

Compared to the centile charts developed by McCarthy et al., (2001), for British WE children and adolescents, SA children in this study at the 50th percentiles (Figures 6.5 & 6.6) had higher WC overall, particularly at the age of 8y onwards in boys and 9y onwards in girls. These differences were much more pronounced at the 90th percentile in both sexes, with SAs having much higher WC measures from age 6y onwards, with slight variations between the sexes of the age at which differences were greatest. Considering that 'adiposity rebound' occurs from around age 5y (Rolland-Cachera et al., 1984), suggests that SA children may have a greater tendency to accumulate abdominal fat at this developmental stage. This aligns well with the IDF (Alberti, 2007) recommendation of determining abdominal obesity in children from age 6y onwards. Based on evidence that overweight and obesity tracks into adulthood (Wardle et al., 2006), targeting preventative measures from age 6y would seem prudent, particularly in this high-risk population.

The centile charts produced by McCarthy et al. (2001), were representative of a WE child and adolescent population from 1988, and similarly the UK90 data was collected between 1978 and 1990 (Cole et al., 1995); with evidence of dramatically increasing levels of obesity in the last 25 years (Bhardwaj et al., 2008; Butland et al., 2007; McCarthy et al., 2003; NHS Information Centre, 2011; WHO, 2011), these comparisons are unlikely to be indicative of ethnic differences between contemporary SA and WE children and adolescents in the UK. Comparisons (based

on SDS values) between a more contemporary sample of WE children and adolescents, using a large dataset of children measured between the years 2004 to 2007 (Table 6.4) at this university, revealed that on average for both genders, SAs had significantly lower ($P<0.001$) BMI and WC than their WE peers. This result would suggest that the SA children would be at lower risk of obesity related diseases than their WE counterparts. However, WC measures do not provide a true indication of visceral fat (Wells & Fewtrell, 2006), and studies comparing WC to abdominal fatness measured by MRI scans have consistently revealed higher correlations with total abdominal fat ($r = 0.8$) than intra-abdominal fat ($r = 0.5$; de Ridder et al., 1992; Fox et al., 1993; Owens et al., 1999). Other studies have reported similar findings, with SA children having lower WC and BMI than their WE counterparts (Nightingale et al., 2011; Whincup et al., 2002). In the study conducted by Nightingale et al. (2011), whilst BMI and WC measures were lower among the SA children compared to the WE cohort, overall levels of adiposity were higher based on BIA and skinfold measure, with higher % fat mass (FM) at an equivalent BMI level.

In terms of prevalence (%) of overweight or obese by BMI, and raised WC (≥90th percentile), no significant differences (at $P<0.05$ level) were observed between both sexes from WE and SA ethnic groups. The prevalence of overweight and obesity determined by the BMI 85th and 95th percentile cut-offs respectively (Table 6.4; NHS, 2011) was far lower than the prevalence of a raised WC percentile close to the Indian adult cut-off at age 16y for 'Action level' 1 or 2 (Misra et al., 2009), or the 90th percentile as proposed by the IDF (Alberti et al., 2007). For instance, at the 95th percentile for BMI, 16.3% of SA boys and 11.2% of SA girls would be classified as obese, compared to 25% of both SA boys and girls having a raised WC ≥90th percentile. In contrast, at the other end of the scale, comparing BMI and WC prevalence at or below the 2nd centile revealed that a greater proportion of both SA and WE boys and girls were categorised below this cut-off compared to WC. This suggests that compared to WC, BMI would overestimate the number of individuals at or below the lower centile cut-offs and underestimate the number of individuals at or above the overweight and obese cut-offs. Based on NICE (2014), guidelines using only the BMI UK90 cut-offs for determining overweight and obesity among all children and adolescents in the UK, would exclude a significant proportion of SAs at

risk of overweight and obesity that would be identified by using the appropriate WC cut-offs.

The sample size for both SA boys and girls aged between 14.00 to 16.99y was severely restricted, which was a limitation of this study. However, all preceding age ranges had large numbers of sufficient power to produce robust curves, and the smoothed centiles produced using the LMS method (Cole and Green, 1992) would account for any skewness in the data. In the study conducted by Ehtisham et al. (2005), the mean WC of the SA adolescents was higher than the mean WC of the SA adolescents aged 14.00-16.99y (n= 19 boys and 12 girls) in this study (79.9cm vs 76.5cm) and (71.9cm vs 67.0cm) for boys and girls respectively. However, the SA population in the study conducted by Ehtisham et al. (2005) had very high levels of overweight or obesity (41% and 42% for males and females respectively), which were significantly higher than the WE population. However, the prevalence of overweight and obesity in this study (24.5% and 21.1%) was similar to the study conducted by Wardle et al. (2006; 21.3% and 29.4%), for boys and girls respectively, with no significant differences found between the SA and WE populations. The 2004 HSE report (Sproston & Mindell, 2006), identified Pakistani boys aged 2-15y as having the highest proportion of overweight or obese children (at 39%) with no other SA ethnicity found to be significantly different to the proportion of overweight and obese children in the general population (at 30% boys and 31% girls).

The significant differences in overweight or obesity, and WC, between the SA and WE cohort in the Ehtisham et al. (2005) study, compared to this and other studies (Wardle et al., 2006), as well as the HSE (2004), may be due to ethnic sub-group differences between SA populations within the UK. In the former study (Ehtisham et al., 2005), participants were recruited from schools in Birmingham and the majority (n=51, 78%) were Pakistani; whilst the study conducted by Wardle et al. (2006), recruited 'Asian' children from schools in London boroughs and did not specify the ethnic sub-group make up. Additionally, the Ehtisham et al. (2005) study had a relatively small sample size (n=65) across a 4y age range, which is likely to have reduced the effect size of the outcome. In this study the majority of participants were Indian (~41%), followed by Bangladeshi (~33%) who were from an area high

deprivation, and Pakistani (~18%), with mixed SA representing the smallest group (~8%). Whilst overall, compared to the WE cohort, SA boys and girls had significantly lower anthropometric measures for all variables (i.e. Ht, weight, BMI, and WC), within-group analyses of SA ethnic subgroups (Table 6.5), identified that Pakistanis were the only group with no significant differences between their WE_2 peers for all measured variables. Furthermore, comparisons revealed that both male and female Pakistanis had significantly higher WC than all other SA ethnic groups. Other studies comparing body composition between SA sub-groups, have similarly reported within-group differences, with Pakistanis reported to have higher levels of obesity than the general population (Sproston & Mindell, 2006). These differences within the SA ethnic group may be attributable to SES, and variations in diet, lifestyle and early feeding practices (Bhopal et al., 1999; Magnhild et al., 2008; Murasko, 2009; Owen et al., 2009; Saxena et al., 2004). Whilst relative risk of diabetes and CVD may vary within SA ethnic sub-groups, there is substantial evidence that the SA ethnicity as a whole is considered a high-risk group (Bodicoat et al., 2014; Nightingale et al., 2011; Whincup et al., 2002; WHO, 2004). Thus based on the evidence from this study and other research, it would be prudent for the previous recommendation by a UK expert group to maintain the BMI status quo across all ethnic groups (Viner et al., 2010), to reconsider this position and revise BMI cut-offs downwards in line with adult cut-offs.

6.4.1 Study Limitations

The SA sample size in the 14-16.99y age range was severely restricted, however, the sample sizes in the preceding age ranges were large enough to reduce the overall impact on the development of the centile curves. In certain age ranges there were also limited numbers of participants from certain SA ethnic sub-groups, to allow for any within-group analyses to be made at individual age ranges. However, for the sample as a whole within-group analyses were conducted to identify any differences between the ethnic sub-groups overall.

6.4.2 Conclusion

Apart from the Pakistani sub-group, SA children and adolescents in this study had significantly lower BMI and WC measures than their WE peers, however in terms of prevalence of overweight and obesity no significant differences were found. The

proportion of children and adolescents identified as overweight or obese determined by BMI was far lower than that determined by WC. This study provides evidence to support the IDF recommendation of using ethnic-specific WC cut-offs for diagnosing metabolic syndrome in children, which is of particular importance in this high-risk population, although neither BMI nor WC per se is likely to provide an indication of overall adiposity within the SA population, and additional measures such as %FM and %skeletal muscle mass from BIA may be required. The results of this study suggest that measuring WC from age 5y onwards would assist in identifying those children at risk of the health consequences associated with abdominal obesity, and enable preventative measures (such as a greater period of time spent in moderate to vigorous exercise) to be adopted as early as possible, based on evidence that obesity tracks into adulthood.

CHAPTER 7: Validation Study of a Field-Based Sitting-Height Measure

7.1 Introduction

Given the significance of total leg length (LL) and LL relative to height (Ht) to future adult health, it is important to have a valid and reliable tool suitable for measuring leg length in field-based studies. Anatomically, LL is defined as the combined length of the femur and tibia (Bogin & Valera-Silva, 2010). However, such measurements can be difficult to conduct in living human subjects, and studies measuring LL have employed different techniques, with some directly taking inside-leg length measurements (BSI, 1990; Gunnell et al., 1998; Pliakas & McCarthy, 2010), or more commonly measuring sitting height (SH; Asao et al., 2006; Bogin & Varela-Silva, 2010; Cameron et al., 1981; Charbonneau-Roberts et al., 2005; Dangour et al., 2002; NHANES, 1988; Torres et al., 2003; Wadsworth et al., 2002), with leg length determined by subtracting SH from standing Ht (figure 1; NHANES, 1988). Although variations in gluteo-femoral fat have been identified as a source of error when estimating leg length using SH; SH appears to be a more popular measure, particularly when measuring overweight or obese people, which is likely due to social and ethical reasons (Bogin & Varela-Silva, 2010).

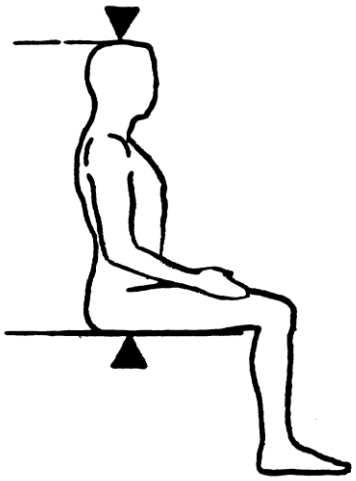


Figure 7.1 Sitting Height position (from NHANES III, 1988)

A SH table, such as the Harpenden SH table (HSHT) fitted with a sliding carriage to adjust to individual thigh length together with an adjustable foot support, is the only

conventional equipment available for measuring SH (Figure 7.2). However, this equipment is bulky, fairly expensive, and non-portable, rendering it impractical for field use. The lack of a standardised tool for measuring SH for fieldwork, has resulted in a variety of methods being employed, from the use of a box, stool or chair placed on or against a stadiometer (Asao et al., 2006; Cameron, 1982; Charbonneau-Roberts et al., 2005; Dangour et al., 2002; NHANES, 1998; Wadsworth et al., 2002; with SH calculated after adjusting where necessary for height of sitting tool), to the use of an anthropometer placed against the subject's back whilst sitting on a table (Cameron et al., 1981; Torres et al., 2003). This latter technique has been described as very difficult to use and prone to error (Cameron, 1982).

Cameron (1982) developed a portable SH tool, which was comprised of a wooden box with an adjustable footboard designed to fit over the base of a Harpenden portable stadiometer. However, this tool is not commercially available and would require considerable expertise and time to construct. Furthermore, it appeared to be a relatively bulky piece of equipment for field use.

7.1.1 Study Aim

The aim of this study was to develop a valid (validated against the HSHT) and more portable SH measure, that would require limited expertise and expense to construct.

Figure 7.2 Harpenden Sitting Height Table image

7.2 Methods

7.2.1 Design of SH tool

A Leicester Height Measure (LHM) was used as the basis for developing the SH tool (Figure 7.3), as it is already comprised of lightweight plastic material which is readily portable. The principle behind the use of the LHM is that it has a platform, which could be adapted to form the sitting area equivalent to that in the HSH table. The platform could then be placed on a firm table and the subject could be measured while sitting on this construction. However, as the base/platform of this stadiometer has raised sides, a piece of lightweight low-density fibreboard (LDF), cut to size at a local DIY store, was obtained at no cost, and fitted into the base to create a

comfortable flat sitting surface (Figure 7.4). The dimensions of the LDF were 2cm (thickness) x 29.5cm (length) x 32cm (width).

Figure 7.3 Leicester Height Measure

Figure 7.4 Adapted Leicester Height Measure

7.2.2 Subject Recruitment

51 volunteers of varying heights and body proportions were recruited for SH measurement. Adult participation was subject to informed consent, and for children, written informed consent was received from the parent/guardian in line with the University's ethical guidelines, with the parent/guardian present during data collection.

7.2.3 Calibration & Measurement procedures

A 1 m wooden rule was routinely used to calibrate the SH measures prior to any measurement of the participants (Figure 7.5). SH was measured by placing the modified stadiometer onto a table and subjects were requested to sit on the table with the back of the knees against the table edge (Figure 7.4). From this position subjects were asked to rise up slightly from the seated position to enable the

modified stadiometer to be correctly positioned beneath the buttocks, ensuring that the backboard was in contact with the sacrum.

Figure 7.5 Calibration of Leicester & Harpenden sitting height measures with 1m rule

Following the procedures recommended by Cameron et al. (1981), SH measurements were taken from the left side of the body. Subjects were requested to sit up tall and straight without leaning the head back against the anthropometer (as this resulted in the head tilting out of position), with feet unsupported and hands resting on the thighs. During initial measurements it was observed that subjects tended to slump a little between measurements which altered results between measures by up to several centimetres, therefore to improve consistency between measures the researcher placed one hand at the base of the spine and another on the left shoulder to draw it back a little, which improved within-subject consistency greatly. The head was placed in the Frankfort Plane position, as described in General Methods (chapter 2 section 2.2.1).

The SH for each subject was measured 3 times on the modified stadiometer and for validation purposes 3 times on a conventional Harpenden Sitting Height Table. The 2.0cm increase in SH, using the modified stadiometer, was corrected for in the recorded results.

7.2.4 Statistical analysis

TEM was assessed including coefficient of reliability as described in Chapter 3 (General Methods section 3.4.1). To validate the modified Leicester stadiometer for SH (LSH) against the SH measures taken on the HSHT (HSH), paired sample t-tests, Pearson's correlation coefficients (using SPSS v.19) and Bland and Altman (1986) plots were carried out.

7.3 Results

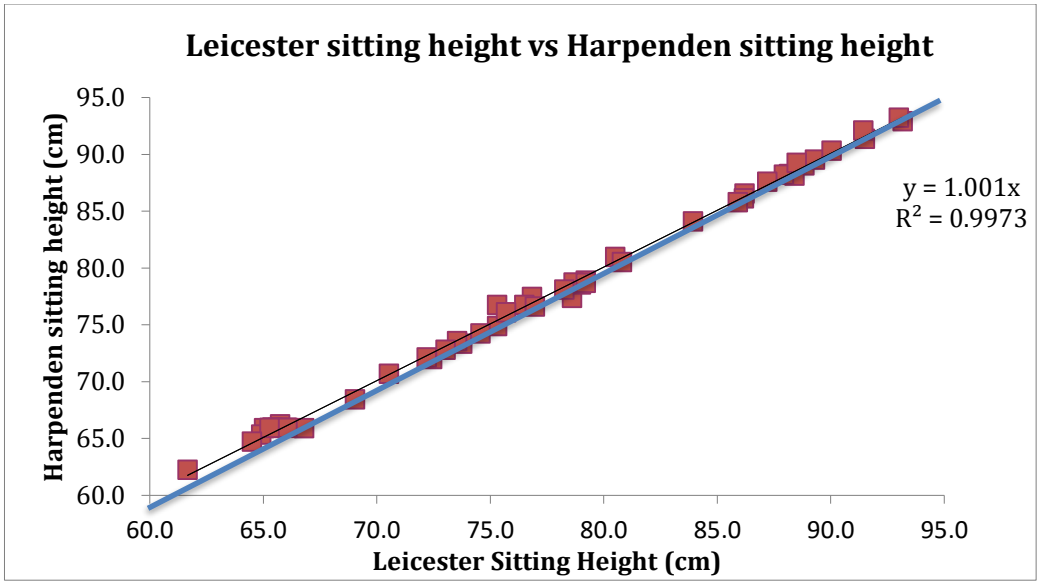
The subject characteristics and SH measures are presented in Table 7.1. Of the 51 subjects measured 25 were boys and 26 girls ranging in age from 5.4y to 22.6y.

Table 7.1 Subject characteristics (age, Ht, LSH, & HSH)

n = 51 (25 boys & 26 girls)		
	Mean	sd (range)
Age (y)	13.1	4.4 (5.4-22.6)
Ht (cm)	149.1	18.0 (117.7-182.0)
LSH (cm)	77.8	9.3 (61.7-93.2)
HSH (cm)	77.8	9.3 (62.2-93.2)

Ht = height; LSH = Leicester sitting height; HSH = Harpenden sitting height

Following preliminary analysis to ensure assumptions of normality were not violated, no significant differences were found between the 2 SH measures t (50) = -1.17, $p > 0.05$ (two-tailed). The mean difference between the measures (LSH – HSH) was -0.1cm with a 95% confidence interval ranging from -0.21 to 0.05. Figure 5 shows a very strong positive correlation between the two measures r = 0.99; $P<0.001$; and Bland-Altman analysis confirmed the very small mean difference of -0.1cm (LSH – HSH) between the 2 methods.



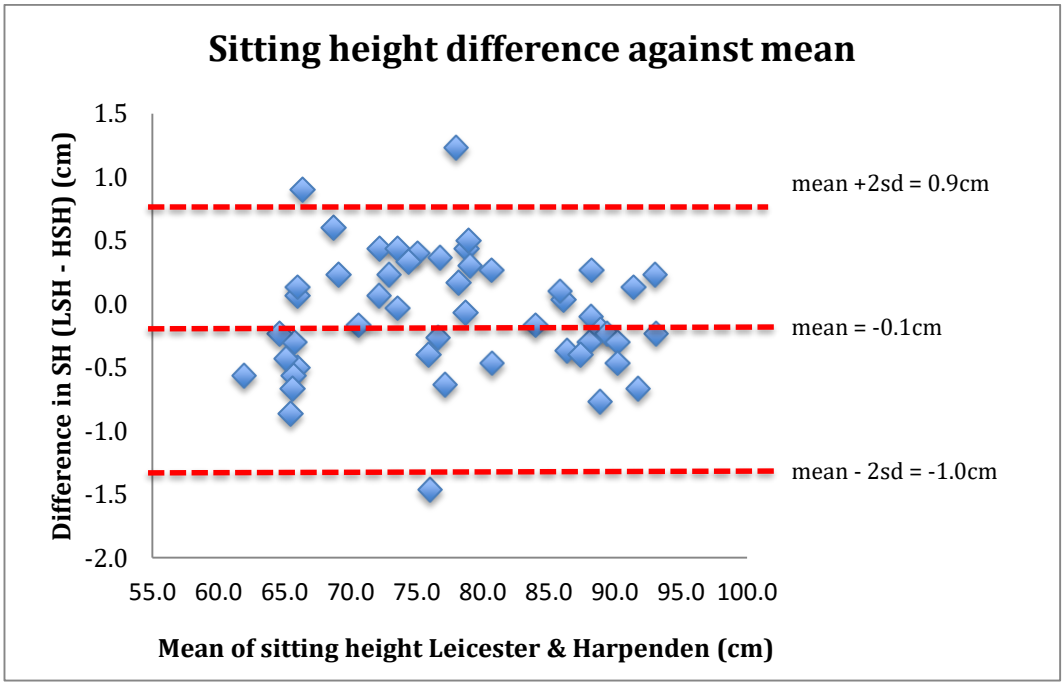


Figure 7.6 Upper graph correlation of LSH vs HSH (r = 0.99, P< 0.001). Lower graph Bland-Altman plot showing limits of agreement between LSH and HSH. LSH = Leicester siting height; HSH = Harpenden sitting height

7.4 Discussion

The aim of this study was to develop and validate a portable tool for measuring SH, as use of previously employed methods, such as taking the inside-leg measure (BSI, 1990; Gunnell et al., 1998; Pliakas & McCarthy, 2010), use of a box (Asao et al., 2006; Cameron et al., 1981), or with subjects sitting on the base-plate of a stadiometer placed on a chair with a flat surface (Wadsworth et al., 2002) was considered impractical, as described in the introduction. The results demonstrate a significant relationship between the portable LSH measure and the HSH ($r = 0.99$), with a mean difference of -0.1cm, and no significant differences between the two measures ($p > 0.05$). As a result of the success of this validation study it was decided that the portable LSH measure would be sufficiently valid to be used in the field-based study to measure the SH of a large South Asian (SA) population of approximately 100 children and adolescents, to compare differences in LL, SH, and relative leg length (RLL) against available data for similar white European (WE) populations.

The limitations of the new LSH measure were that it was not as rigid as the HSHT, as the upright rule did not provide as firm a support to the back, which was much narrower than the backboard of the HSHT. This resulted in having to adjust participants' seating position to ensure the back was kept straight. A firm, stable, and steady table was required for placement of the LSH measure that would support the weight of participants, although this would normally be available in most school and clinic settings. In conclusion, the adjusted LSH measure developed in this study is a suitable field-based measure for SH as it is lightweight, portable, accurate and precise.

CHAPTER 8: A comparison of leg length and stature between South Asian & white European children & adolescents

8.1 Introduction

Chapter 2 (section 2.4) provides an overall review of the literature on stature and body proportionality, in particular relative leg length (LL) determined by the leg length to height (Ht) ratio (LLHR; LL/Ht) or sitting height (SH) ratio (SHR; SH/Ht). Short stature in adulthood is associated with an increased risk of obesity and metabolic syndrome (Guerrero-Igea et al., 2001; Gunnell et al., 2003; Lawlor et al., 2002); in contrast, taller for age children have been shown to have a greater propensity to obesity (Baker et al., 2007; Bosy-Westphal et al., 2009; Buchan et al., 2007; Demerath et al., 2009; Himes et al., 1986; Lazarus et al., 1996), which has been related to advanced sexual maturity among girls (Adair et al., 2001; Freedman et al., 2003). Additionally, shorter LL, particularly, relative LL, is considered an increased risk factor for type 2 diabetes mellitus (T2DM) and cardiovascular disease (CVD) as it is associated with higher levels of adiposity (Asao et al., 2006; Bogin & Varela-Silva, 2010; Pliakas & McCarthy, 2010; Hales & Barker, 2001; Smith et al., 2001; Wadsworth et al., 2002).

Population differences in stature, weight and overall body proportionality, have been attributed to a combination of genetic and environmental factors (including diet and nutrition; Eveleth & Tanner, 1990; Rona & Chinn, 1986), although in healthy populations, the environment is considered more influential in determining stature and LL than genetics (Bogin & Valera-Silva, 2010), as demonstrated by evidence of secular increases in these body proportions (Bogin et al., 2002; Bogin & Valera-Silva, 2010; Tanner et al., 1982; Wadsworth et al., 2002). In a study conducted by Hauspie et al. (1996), comparing the heights of children from West Bengal measured between 1952-1956, to the heights of children from the UK, found that the mean heights of the Bengali children fell below the 10th centile of the UK references. In The National Study of Health and Growth (Rona & Chinn, 1987), comparing the growth of different ethnic groups within the UK, Ht was strongly associated with ethnicity of children living within inner city areas. Within this study, children of South Asian (SA) origin were the shortest, whilst African-Caribbean children were the tallest.

Kelly et al. (1997), conducted a study comparing the growth of 785 Pakistani children aged 5-14 years, from a deprived area of Birmingham, to the UK90 growth standards for stature and weight (Freeman et al., 1995), body mass index (BMI; Cole et al., 1995), and SH and subischial LL constructed in the 1970s (Tanner & Whitehouse, 1978). In that study, compared to white European (WE) children, Pakistani children of both sexes were on average shorter and lighter (Pakistani boys: mean Ht < 0.2SD; mean weight (Wt) < 0.3SD; Pakistani girls: mean Ht < 0.4SD; mean Wt <0.5SD), and had lower BMIs (Boys overall difference = -0.4SD; girls overall difference = -0.5SD). These differences varied by age, with mean differences for each age group falling between the 25th and 50th centiles. Differences in SH and LL between the Pakistani children and the WE UK reference data (Tanner & Whitehouse, 1978) were more marked, with SH ranging between the mean and mean -1SD for boys and girls; -2SD for girls at 13-14 years only with corresponding LL falling to the mean; and LL ranging between mean and mean + 1SD. Whilst it appears from that study that Pakistani children were on average shorter and lighter than WE children, their body proportions also appeared to be quite different, with longer legs compared to SH. However, it is important to note that the LL and SH data were compared to charts constructed in the 1970s. The authors commented that the differences in the latter measures were more likely to be due to difference in methodology rather than actual growth differences. Similar differences in RLL were reported by Dangour et al. (2002), when comparing a more contemporary cohort of WE children, to the British reference curves for SH and LL, developed by Tanner and Whitehouse (1978).

Low SES and adverse environmental conditions are reported to have a detrimental impact on growth (Bogin & Varela-Silva, 2010; Martin et al., 2002; Tanner et al., 1982; Wadsworth et al., 2002), thus differences in stature, weight, and BMI in the study by Kelly et al. (1997), may be attributed more strongly to environmental conditions, as these children were described as being from "one of the most deprived areas of Western Europe" (p.410). Furthermore, longitudinal studies have shown that children of Pakistani origin from more affluent backgrounds living in the USA have very similar growth patterns to the general US population, when

compared to the growth curves of the NCHS (Hamill et al., 1979), although others have reported a greater divergence worldwide in post-adolescent Ht across population groups from high SES backgrounds (Eveleth & Tanner, 1990; Haas & Campirano, 2006), with Europeans being considerably taller than non-Europeans.

Growth during infancy and childhood is highly plastic (Bogin & Valera-Silva, 2010; Hales & Barker, 2001), with adverse environmental conditions such as poor foetal or infant nutrition (Asao et al., 2006; Wadsworth et al., 2002), and low SES directly affecting stature, particularly limb length (Bogin & Varela-Silva, 2010; Wadsworth et al., 2002). However, in population groups from high SES backgrounds, according to Eveleth and Tanner (1990), whilst post-adolescent Indian children were shorter than their WE counterparts, they had similar relative SH, indicating similar RLL. Other studies have reported similar findings such as a prospective study conducted by Krishnaveni et al. (2005), on Indian children from birth to 4y (see Chapter 2 Literature Review section 2.6), where the Indian children compared to WE children were considerably smaller, but had a longer RLL. In a recent cross-sectional study conducted in New Zealand, where dual-energy x-ray absorptiometry (DXA) was used to compare body size, body composition, and fat distribution, between European, Maori, Pacific Island, and Asian Indian (which included people of Pakistani, Indian, and Sri Lankan origin) adults (n= 933; age = 17-80y), whilst the Asian Indians had more body fat, both total and abdominal, with less fat-free mass (FFM), skeletal muscle mass (SMM) (including appendicular SMM (SMM_a)), and bone mineral, than the other ethnic groups, compared to Europeans RLL was similar for men but longer for women (Rush et al., 2009). However, both Asian Indian men and women had the same %FM at lower BMIs (24kg/m^2 and 26kg/m^2 respectively) than Europeans with a BMI of 30kg/m^2, or Pacific Island men and women with a BMI of 34 kg/m^2 and 35 kg/m^2 respectively, highlighting the inadequacy of BMI for the SA population.

In 2002, Dangour et al. developed up to date references for SH and LL for contemporary white British children measured between 1995-1996, to replace the Tanner and Whitehouse (1978) reference curves. Currently, there are no references for SA children in the UK.

8.1.1 Study Aims

The aims of this study were:

i) To compare ethnic differences in LL, RLL, and stature between SA and WE children and adolescents in the UK.

ii) To review the health implications, if any, of these differences.

8.2 Methods

8.2.1 Participants and Recruitment Procedures

Children and adolescents recruited for the bioelectrical impedance analysis (BIA) validation study for body composition (Chapter 4) were also requested to participate in measurements for SH and waist circumference (WC), which was included in the participant consent forms. A total of 171 healthy children and adolescents aged 5-18y, recruited from various community centres and mosques in Greater London, were directly measured (108 boys and 63 girls). Participants were requested to specify ethnicity on their consent forms, and those that had parents from more than one SA ethnic group (e.g. Indian and Pakistani) were defined as mixed SAs. All measures were taken following participant consent as explained in the recruitment procedures in Chapter 3 (General Methods).

8.2.2 Anthropometric Methods

Ht was measured using a portable Leicester Height Measure (LHM), and following the successful modification of the LHM, as detailed in chapter 6, SH was measured. Wt and WC measures were also taken. Detailed methodology is provided in chapter 3 for all measurements taken (General Methods). A single observer took all measures, to prevent inter-observer error. Technical Error of Measure (TEM) and coefficient of reliability (R) was determined as explained in General Methods (chapter 3).

8.2.3 Statistical Methods

Standard deviation scores (SDS) were generated in Excel, for all anthropometric variables including BMI and WC as described in General Methods (Chapter 3 section 3.4.2). Data was analysed using Microsoft Excel 2011 and SPSS software version 22. Data are presented as means ±SD. Sex-specific between group (SA and WE) and sex- and ethnic-specific (i.e. Indian, Pakistani, Bangladeshi, and mixed SA) within- and between-group (i.e. specified SA ethnicity and WE) comparisons for all

anthropometric measures presented in SDS format were conducted on the whole sample and also split by age range 5-11y and 12-18y, for pre- and post-pubertal comparisons, using independent samples t-tests and one-way ANOVA with Bonferroni post-hoc tests. Bivariate comparisons for stature (Ht-SDS) and LLHR, with SDS values for WC and BMI using Pearson's product-moment correlations in SPSS were also conducted.

8.3 Results

Data from a total of 171 (108 boys and 63 girls), aged 5-18y were obtained. Descriptive data for the whole sample (separated by sex) is shown in Table 8.1, providing anthropometric characteristics presented as SDS values, based on UK90 growth reference data. LLHR and SHR are also presented. Compared to the UK90 reference data, both SA boys and girls had lower than average SDS values for Ht, Wt, BMI, and SH. However, LL was longer (SDS 0.21) for boys, and very slightly longer (SDS 0.05) for girls. WC on the other hand was considerably higher (SDS 0.67, and SDS 0.65 for boys and girls respectively), than the UK90 reference population. No significant ($P<0.05$) differences were observed between the sexes for any of the measured anthropometric variables as presented in Table 8.1 following t-tests.

Table 8.1 Anthropometric characteristics (Ht, Wt, BMI, WC, SH, LL absolute and SDS, & % LLHR & SHR) of SA cohort by sex

	Boys (n =108)		**Girls** (n = 63)	
	Mean	±sd	Mean	±sd
Age (y) (range)	10.2 (5.3-18.9)	3.32	11.0 (5.1-18.9)	3.57
Ht SDS	-0.07	0.97	-0.22	1.14
Wt SDS	-0.11	1.25	-0.23	1.16
BMI SDS	-0.17	1.38	-0.23	1.34
WC SDS	0.67	1.26	0.65	1.23
SH SDS	-0.52	0.95	-0.53	1.13
LL SDS	0.21	0.97	0.05	1.13
LLHR (%)	47.96	1.61	47.50	1.45
SHR (%)	52.03	1.61	52.50	1.45

Ht = height; Wt= weight; BMI = body mass index; WC = waist circumference; SH= sitting height; LL = leg length; LLHR = leg-length-to-height-ratio LL/Ht; SHR = sitting-height-ratio SH/Ht; SDS = standard deviation score; SA = South Asian.

Table 8.2 shows the sex-specific anthropometric characteristics (both absolute and SDS) of the cohort, separated into two age-ranges (<12y and ≥12y), to identify any differences in body proportionality, that may occur pre- and post-puberty. T-tests were conducted to compare differences in anthropometric variables (all SDS data, LLHR, and SHR) between the same sex groups pre- and post-puberty (i.e. <12y and ≥12y), and between the sexes within the two age ranges. For boys, the pre-pubertal group had a significantly (P<0.001) lower LLHR and conversely higher SHR than post-pubertal boys, as well as a significantly (p= 0.008) higher SDS (SDS 0.85 vs SDS 0.07) for WC. For girls, based on UK90 SDS values, the post-pubertal girls were significantly (p=0.037) shorter (SDS -0.65 or 26th centile), with a lower LL (p= 0.02; SDS -0.42 than the pre-pubertal girls (SDS -0.01), but had a significantly higher BMI (p= 0.001; SDS 0.53 vs SDS -0.62), and WC (p= 0.043; SDS 1.16 vs SDS 0.45). Comparisons between the sexes in the pre-pubertal age group, revealed only significant (p=0.046) differences for SDS BMI, with girls having a significantly lower BMI (SDS -0.62) than boys (SDS -0.10).

Compared to the UK90 reference population in general, the pre-pubertal boys were far more similar to the reference population for Ht, Wt, and BMI (52nd centile; 46th centile; 42nd centile respectively) than the post-pubertal group, who were much shorter, lighter, with a lower BMI (37th centile; 38th centile; 37th centile respectively). Both pre- and post-pubertal boys had on average a larger WC and longer LL than the reference population, however, the pre-pubertal boys had a much larger SDS value for WC (80th centile vs 53rd centile), and a comparatively longer LL (60th centile vs 53rd centile). The pre-pubertal girls were also much closer (although slightly shorter) in Ht to the reference population (47th centile) than the post-pubertal girls who were much shorter on average (26th centile). However, the pre-pubertal girls had a much lower average Wt (32nd centile), and BMI (25th centile), whilst the post-pubertal girls were on average heavier (54th centile), with a much higher BMI (70th centile) than the reference population. The pre-pubertal girls as with pre-pubertal boys, also had on average a longer LL (61st centile); the post-pubertal girls had on average a shorter LL (32nd centile), although SH was even lower (19th centile), indicating a greater LLHR than the reference population.

To compare relative LL of the UK90 reference population to the SAs in this study for pre- and post-pubertal boys and girls, the mean stature for each sex from 5-11y and 12-18y was summed and divided by the mean LL for each age range, and then converted to a percentage (i.e. multiplied by 100) LLHR. The LLHR values computed were 46.7% and 47.9%, and 46.7% and 47.0% for pre- and post-pubertal boys and girls respectively. This equated to SAs having a longer LLHR by 0.9% and 1.2%, and 0.8% and 0.5% for pre-and post-pubertal boys and girls respectively.

Table 8.2 Anthropometric characteristics (Ht, Wt, BMI, WC, SH, LL absolute and SDS, & % LLHR & SHR) of SA cohort by sex and age-range including pre- and post-pubertal comparisons

	Pre-pubertal				**Post-pubertal**			
	Boys (n =78)		**Girls** (n = 42)		**Boys** (n=30)		**Girls** (n=21)	
	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD
Age (y) (range)	8.5 (5.3-11.7)	1.81	8.9 (5.1-11.9)	1.95	14.60 (12.0-18.9)	2.13	15.13 (12.0-18.9)	2.81
Ht (m)	1.30	0.10	1.32	0.12	1.60	0.13	1.56	0.07
Wt (kg)	28.4	8.15	28.31	8.50	50.12	16.15	53.70	12.25
BMI	16.4	2.88	15.85	2.65	18.96	3.84	22.00	4.21
WC (cm)	61.00	8.54	58.64	7.88	70.06	10.64	71.89	10.00
SH (cm)	68.3	4.50	69.23	5.45	81.65	7.46	81.89	4.35
LL (cm)	62.0	6.42	62.93	7.35	78.83	6.13	73.97	3.32
LLHR (%)	47.5**	1.58	47.53	1.56	49.15**ΔΔ	0.92	47.47ΔΔ	1.16
SHR (%)	52.5**	1.58	52.47	1.56	50.85**ΔΔ	0.92	52.53ΔΔ	1.16
Ht SDS	0.04	0.94	-0.01*	1.05	-0.36	0.98	-0.65*	1.22
Wt SDS	-0.00	1.38	-0.41	1.10	-0.39	1.26	0.11	1.21
BMI SDS	-0.10Δ	1.38	-0.62*Δ	1.29	-0.35Δ	1.40	0.53*	1.13
WC SDS	0.85*	1.24	0.45*	1.12	0.07*Δ	1.18	1.16*Δ	1.38
SH SDS	-0.42	0.91	-0.38	1.10	-0.78	1.01	-0.83	1.17
LL SDS	0.26	0.99	0.28*	1.13	0.07	0.89	-0.42*	0.99

*Significant difference between same sex groups <12y & ≥ 12y at p <0.05 level; ** Significant difference between same sex groups <12y & ≥ 12y at p <0.001 level; ΔSignificant difference between sexes at <12y & ≥ 12y at p <0.05 level ΔΔSignificant difference between sexes at <12y & ≥ 12y at p <0.001 level; Ht = height; Wt= weight; BMI = body mass index; WC = waist circumference; SH= sitting height; LL = leg length; LLHR = leg-length-to-height-ratio LL/Ht; SHR = sitting-height-ratio SH/Ht; SDS = standard deviation score; SA = South Asian.

Pearson's correlations were conducted to assess the relationship of WC and BMI SDS values with stature (Ht SDS) and relative LL (LLHR). This test revealed that both Ht SDS and LLHR were significantly correlated with WC SDS (r= 0.292; P<0.001 and r=-

0.179; p= 0.026 respectively) and BMI-SDS (r=0.177; p= 0.022 and r= -0.186; p= 0.016 respectively). The positive correlation with SDS-Ht indicated that as Ht increased relative to the reference population (UK90), there was a concomitant increase in BMI and WC SDS. However, the negative correlation with LLHR indicated that as relative LL increased, BMI and WC decreased in comparison to the UK90 reference population.

Further sex-specific comparisons were made for Ht SDS, LLHR and SHR between the top and bottom quartiles (i.e. SDS≤ -0.61 and ≥ 0.66 respectively) for BMI and WC SDS (Tables 8.3 and 8.4 respectively). For boys, compared to the UK90 reference population, individuals in the bottom SDS quartiles for BMI and WC were shorter, whilst those in the top SDS quartiles were taller. Between-group comparisons revealed differences to be significant (p= 0.003 and 0.028 for BMI and WC SDS respectively). However, whilst overall LL SDS was also lower (between-group) for BMI and WC SDS in the 'Low' BMI and WC SDS groups, they had relatively longer legs (i.e. greater %LLHR), although this difference was only significant in the 'Low' WC SDS group (p= 0.007), with a conversely shorter SH, than individuals in the top quartiles. For girls however, for overall height, individuals in both the 'High' and 'Low' BMI SDS groups, relative to the UK90 reference population, were shorter; however, those in the 'High' BMI SDS group were even shorter than those in the 'Low' BMI SDS group, although the between-group differences were not significant. Those in the 'Low' BMI SDS group on the other hand had a significantly (p= 0.014) greater LLHR. For WC SDS, girls in the bottom quartile were on average shorter than those in the top quartile, however, their relative LL (LLHR) was longer, although this difference was not significant.

Table 8.3 Sex-specific differences mean ± SD between SA cohort and UK90 references in Ht SDS, RLL and SH between 'Low' and 'High' BMI SDS

	Boys		Girls	
	Low BMI SDS (n=40)	High BMI SDS (n=32)	Low BMI SDS (n=27)	High BMI SDS (n=18)
	Mean ±SD	Mean ±SD	Mean ±SD	Mean ±SD
Ht SDS	-0.44* ±0.87	0.25* ±1.05	-0.28 ±1.07	-0.36 ±1.31
LL SDS	-0.05 ±0.85	0.33 ±1.08	0.21 ±1.10	-0.35 ±1.10
LLHR (%)	48.13 ±1.74	47.65 ±1.49	47.99* ±1.43	46.93* ±1.27
SHR (%)	51.87 ±1.74	52.35 ±1.49	52.01* ±1.43	53.07* ±1.27

*Significantly different (within-group) between 'Low' and 'High' BMI SDS groups at P<0.05 level

SA = South Asian; BMI = body mass index; Ht = height; LL = leg length; LLHR = leg-length-to-height-ratio LL/Ht; SHR = sitting-height-ratio SH/Ht; SDS = standard deviation score. 'Low' & 'high' BMI SDS = bottom & top SDS quartiles respectively for BMI

Table 8.4 Sex-specific differences mean ± SD between SA cohort and UK90 references in Ht SDS, RLL, and SH between 'Low' and 'High' WC SDS

	Boys		Girls	
	Low WC SDS (n=14)	High WC SDS (n=48)	Low WC SDS (n=11)	High WC SDS (n=30)
	Mean ±SD	Mean ±SD	Mean ±SD	Mean ±SD
Ht SDS	-0.42* ±0.72	0.22* ±0.99	-0.44 ±0.81	0.02 ±1.13
LL SDS	0.07 ±0.74	0.37 ±1.09	0.00 ±1.12	0.10 ±1.15
LLHR (%)	48.85* ±1.34	47.58* ±1.67	47.44 ±1.07	47.23 ±1.31
SHR (%)	51.15* ±1.34	52.42* ±1.67	52.56 ±1.07	52.76 ±1.31

*Significantly different (within-group) between 'Low' and 'High' WC SDS groups at P<0.05 level

SA = South Asian; WC = waist circumference; Ht = height; LL = leg length; LLHR = leg-length-to-height-ratio LL/Ht; SHR = sitting-height-ratio SH/Ht; SDS = standard deviation score. 'Low' & 'high' WC SDS = bottom & top SDS quartiles respectively for WC

Table 8.5 provides descriptive data for the sample by ethnic sub-group. As there were only 3 individuals altogether in the 'Other' SA category respectively, this data is not shown. Furthermore, there were only 11 Pakistani and 5 Bangladeshi girls, significantly reducing the statistical power of any tests. However, this data was still retained for analysis. Comparisons between absolute values for anthropometric measures were not made, as the mean ages between the ethnic sub-groups differed, particularly the Pakistani sub-group, with a lower mean age and smaller age range. However, sex-specific between sub-group comparisons for LLHR and SHR, as well as for all SDS data were conducted. In terms of relative leg length (LLHR), all sub-groups had similar ratios with no significant differences. For both boys and girls, the Pakistani sub-group were the only group found to be taller on average than the

UK90 reference population (59th centile), although between-group differences were not significant. Both Indian and Bangladeshi sub-groups were shorter than the UK90 population, although girls were relatively shorter than boys in comparison to the reference population (45th and 44th centile vs 36th and 38th centile respectively). However, the Bangladeshi girls were the only sub-group to have a shorter LL (32nd centile) than the reference population, although as already stated the between sub-group differences for RLL were not significant. For BMI SDS there were no between sub-group differences for boys, although the Bangladeshi sub-group had a higher BMI than the reference population (0.40 SDS), whilst the Pakistani sub-group were very close to the population average (0.01 SDS), and Indians had a lower BMI on average (-0.44 SDS). Bangladeshi girls had a significantly higher BMI than both Indian and Pakistani girls; however, the Bangladeshi and Pakistani sample size was very small.

Table 8.5 Sex-specific anthropometric (Ht, Wt, BMI, WC, SH, LL SDS, & % LLHR & SHR) comparisons between SA ethnic sub-groups

	Boys			Girls		
Ethnic code	1 (n=59)	2 (n=27)	3 (n=21)	1 (n=45)	2 (n= 11)	3 (n=5)
δ Age (y) (range)	11.00 (5.4-18.9) (3.35)	8.11 (5.3-11.4) (1.82)	10.70 (5.8-18.2) (3.73)	11.23 (5.6-18.9) (3.65)	8.62 (5.1-11.7) (2.29)	14.1 (10.5-17.9) (3.03)
LLHR (%)	48.2 (1.48)	47.63 (1.8)	47.82 (1.68)	47.62 (1.50)	47.19 (1.43)	47.16 (1.08)
SHR (%)	51.8 (1.48)	52.37 (1.80)	52.18 (1.68)	52.38 (1.50)	52.82 (1.43)	52.83 (1.08)
Ht SDS	-0.17 (0.92)	0.22 (0.98)	-0.15 (1.05)	-0.33 (1.22)	0.21 (0.79)	-0.38 (1.21)
β Wt SDS	-0.33 (1.21)	0.14 (1.20)	0.21 (1.40)	-0.49* (1.00)	-0.06* (1.28)	1.44* (1.03)
β BMI SDS	-0.44 (1.39)	0.01 (1.17)	0.40 (1.50)	-0.47* (1.17)	-0.33* (1.57)	1.84* (0.79)
β WC SDS	0.33* (1.28)	1.04 (1.02)	1.23* (1.24)	0.39** (0.96)	0.57** (1.37)	3.10** (0.60)
SH SDS	-0.60 (0.93)	-0.37 (1.07)	-0.46 (0.86)	-0.67 (1.21)	-0.08 (0.63)	-0.48 (1.24)
LL SDS	0.10 (0.86)	0.54 (1.01)	0.11 (1.13)	0.00 (1.21)	0.42 (0.95)	-0.42 (0.87)

Data are means (±SD); δ For 'Age', mean age and age range (in brackets) is presented in addition to ±SD (in brackets). *Significantly different at the p <0.05 level; **Significantly different at the p <0.001 level; SA = South Asian; ethnic codes: 1= Indian; 2= Pakistani; 3= Bangladeshi; βWithin group differences for Wt, BMI, and waist circumference for girls were between Indian and Bangladeshi and Pakistani and Bangladeshi sub-groups only. Ht = height; Wt = weight; BMI = body mass index; WC = waist circumference; SH = sitting height; LL = leg length; LLHR = leg-length-to-height-ratio LL/Ht; SHR = sitting-height-ratio SH/Ht; SDS = standard deviation score.

8.4 Discussion

There is much evidence to support the association of taller for age children and short stature in adulthood, and a shorter relative leg length, with obesity and its related diseases, such as T2DM and CVD (Asao et al., 2006; Bogin & Varela-Silva, 2010; Bosy-Westphal et al., 2009; Buchan et al., 2007; Pliakas & McCarthy, 2010; Hales & Barker, 2001; Smith et al., 2001; Wadsworth et al., 2002). Dangour et al. published up to date reference data on Ht, SH and LL for WE children and adolescents in 2002. However, there is no contemporary data available of SH and LL for SA children living in the UK. This study fills an important gap in the research by providing up to date evidence of the differences in body proportionality between SA and WE children and adolescents in the UK, and how body proportionality, in particular stature and RLL relates to measures of obesity such as BMI and WC. These findings need to be

interpreted with caution however, as the sample was opportunistic and relatively small, and thus could not be regarded as representative of the whole UK SA population.

Due to a relatively modest sample size, age-specific analyses for each year group were not conducted. However, the sample was split in half by age range to provide an indication of any pre- and post-pubertal differences in growth relative to the UK90 reference population (Dangour et al., 2002), although actual age of puberty was not determined. This study revealed that overall both SA boys and girls were on average shorter, lighter, with a lower BMI and SH, but had a much higher WC, and longer LL. The marked differences in BMI and WC between the SA and WE samples add to the body of evidence that BMI, particularly for SAs, is not a reliable indicator of obesity (Ehtisham et al., 2005; Nightingale et al., 2011; Rush et al., 2009; Saxena et al., 2004; Viner et al., 2010).

Whilst the SAs were on average shorter than the UK90 population, a lower SH and longer LL, would indicate that SAs have relatively longer legs than their WE counterparts. This finding has been observed elsewhere, where SA children even from more deprived backgrounds than this cohort that were shorter and lighter, had comparatively longer legs and a shorter SH than WE children (Kelly et al., 1997; Krishnaveni et al., 2005). However, in the study conducted by Kelly et al. (1997), this outcome may have been attributed to differences in measurement methods. Eveleth and Tanner (1990) also observed that affluent Indians compared to WEs were on average shorter, but had a shorter SH and similar relative SH, indicating a similar RLL. In the study conducted by Rush et al. (2009), whilst the WE cohort were taller than the SAs, for men RLL was similar, and SA women had a longer RLL, although appendicular SMM was significantly lower than all the other ethnic groups. As a proportion of total fat, SAs had the highest abdominal fat, whilst WEs had the lowest abdominal fat but the highest thigh fat in that study.

Comparisons between pre- and post-pubertal measures revealed that both pre-pubertal boys and girls were much closer in Ht to the reference population, but post-pubertal heights were much lower. This indicates that the peak height velocity or

rate of growth during the adolescent growth spurt was much slower in the SA cohort than in the WE reference population, resulting in a shorter adult Ht. A similar divergence in post-pubertal heights between non-European populations and WE has also been reported in reviews (Eveleth and Tanner, 1990; Haas & Campirano, 2006).

As the UK90 data (Dangour et al., 2002), did not provide any reference measures for relative LL (i.e. SHR or LLHR), it was difficult to make these comparisons with our study population, a factor that has also been commented on recently (Bogin & Valera-Silva, 2010). To overcome this difficulty, mean data was used to calculate LLHR and convert to a percentage. From this analysis it was revealed that both SA boys and girls had on average relatively longer legs than WEs, ranging from 0.5% to 1.2%.

The relationship between BMI and WC with stature and RLL, revealed a significantly positive relationship with Ht for both BMI and WC, and a significantly negative relationship with LLHR for both these measures. This indicated that taller stature in childhood had a positive association with measures of obesity, and conversely a relatively longer leg length had a negative association, which is consistent with the findings of other research (Asao et al., 2006; Bogin & Varela-Silva, 2010; Bosy-Westphal et al., 2009; Buchan et al., 2007; Pliakas & McCarthy, 2010; Hales & Barker, 2001; Smith et al., 2001; Wadsworth et al., 2002). Further sex-specific comparisons between RLL (i.e. LLHR and SHR) and Ht SDS, with the top and bottom quartiles for BMI and WC SDS values revealed a negative association between these measures and RLL for both sexes, i.e. those in the top quartiles for BMI and WC had a shorter RLL than those in the bottom quartile. However, Ht SDS comparisons varied by sex, with boys in the top BMI and WC SDS quartiles being significantly taller, whilst girls were shorter (although not significantly), than their sex-matched counterparts in the bottom BMI and WC SDS quartiles.

The sex-specific sub-group comparisons were limited in statistical power due to a reduced sample size, particularly within the female sub-group, that had only 11 Pakistani, and 5 Bangladeshi girls. Secondly, as the Pakistani sub-group had a lower

mean age range, comparisons between absolute values were not made. Comparisons between the UK90 reference population revealed that the Pakistani sub-group were the only group that were taller; however, as the age range for this sub-group fell within the pre-pubertal age range, may be a contributory factor, based on the analyses between the pre- and post-pubertal groups as stated above.

8.5.1 Study Limitations

As already stated the sample size was too small to make any analyses by individual year group. Ethnic-specific sub-group analyses were also limited due to small sample sizes, particularly for girls in the Bangladeshi and Pakistani sub-groups. The UK90 reference standards (Dangour et al., 2002) represent children and adolescents measured in 1995-1996, and thus may not now be representative of a more contemporary WE population, based on evidence of secular increases in Ht. Furthermore, UK90 data for RLL was not available, and had to be derived. More up to date references with RLL data would be required to confirm the findings of this study, and SES would need to be controlled for, based on evidence that it impacts stature and RLL. Whilst assessment of TEM was used to minimise intra-observer error, inter-observer differences when making comparisons between studies are not controllable. Therefore, to minimise inter-observer error, it is vital that strict standardised measurement protocols are followed.

8.5.2 Conclusion

According to the findings of this study SA children and adolescents have relatively longer legs than their WE counterparts. Pre-pubertal girls and boys have similar overall stature to their WE counterparts; however, adolescent SAs tend to be considerably shorter than WEs. This study confirms the findings of other research which shows that relatively shorter legs and taller for age stature in childhood is positively associated with measures of obesity such as BMI and WC. The well-recognised associations of diabetes and CVD, with increased abdominal adiposity and reduced skeletal muscle mass among SAs may be slightly offset by their relatively longer legs, although this finding would need to be qualified with more direct between-group comparisons, as RLL for the UK90 population was not available and had to be derived. Increasing muscle mass in the lower limbs may help this high-risk population group to reduce the risk of developing obesity related

diseases, although further research would be required to investigate how modifiable SMMa is, given that SAs have less appendicular SMM than other ethnicities. While the work in this chapter has highlighted an area of human biology where there may be ethnic differences, much more work needs to be carried out to verify these findings, especially in larger samples and different SA groups across the UK.

CHAPTER 9: Conclusion

9.1 Overall Aims of Thesis

The overall aims of this thesis were to:

i) Validate a tetrapolar bioelectrical impedance analyser (BIA; Tanita BC418-MA) for body composition assessment (BCA) using dual-energy x-ray absorptiometry (DXA) and air displacement plethysmography (ADP) as the reference methods for the South Asian (SA) child and adolescent population.

ii) Develop body composition (BC; %fat mass (FM) and appendicular skeletal muscle mass (SMMa) measures) and waist circumference (WC) reference centiles for this population group.

iii) Validate a sitting height (SH) measure for field-based use for the determination of relative leg length (RLL) and SH.

iv) Examine ethnic differences in BC, WC, and body proportionality in terms of stature and RLL between this population group and their white European (WE) peers, as well as examine any within-group differences.

9.2 Summary of Research and Findings

9.2.1 BIA validation study – Chapter 4

Prior to application of the new BIA prediction equation (BIA_{reg1}) developed in this study, BIA compared to DXA was significantly underestimating %FM. BIA_{reg1} reduced the mean differences between BIA and DXA in both boys and girls. The equation developed in this study demonstrated an improvement in outcome for estimating fat-free mass (FFM) when compared to a similar study where the same BIA model was validated against DXA. A limitation of BIA observed in this study and others, is that it has a tendency to overestimate %FM at the leaner end of the scale and underestimate %FM at high adiposity levels. This tendency was reduced when the new equation developed in this study was applied. The validity of BIA_{reg1} was also assessed, by applying the new equation to a large external SA dataset of children

and adolescents, which found the equation was only valid for children aged ≥9y (see section 9.3 Limitations).

Comparisons were also made between BIA and DXA for SMM_a, and whilst correlations for both absolute (kg) and relative (%) SMM_a between the two methods were strong, the average differences were significant but small with BIA underestimating SMMa. However, as DXA weight (Wt) was not equivalent to scale Wt and required adjustment, it was not possible to adjust for limb Wt. Therefore, no correction factor was applied for SMM_a measures, and whilst the differences were statistically significant, as they were very small in reality, original BIA values were used in the analyses. Furthermore, as DXA Wt was greater than scale Wt and BIA was found to overestimate total body FM, if SMM_a could be corrected, it would more likely reveal that BIA overestimated this measure.

9.2.2 Ethnic differences in body composition and development of ethnic specific reference centiles – Chapter 5

Anthropometric SDS comparisons of UK90 reference data for height (Ht, Wt, body mass index (BMI), WC, and %FM with the SA cohort revealed that SA boys and girls were on average shorter, lighter, with a lower BMI, but with a higher WC and %FM, which highlights the inadequacy of BMI as a reliable indicator of adiposity and supports the considerable body of evidence that other more direct measures of adiposity are required in addition to BMI, particularly in this high risk population.

Similar comparisons were also made with the SA cohort and a more contemporary low income WE_2 sample. Results revealed that compared to the SA sample, the WE_2 cohort had higher mean SDS for all measures. Comparisons between the SA and WE_2 cohort, revealed that both SA boys and girls were significantly shorter, lighter, with a lower BMI and WC than their WE_2 counterparts, with no significant differences in %FM. The WE_2 results illustrate the secular increase in standard anthropometric measures, particularly BMI, %FM, and WC. However, for the SA cohort whilst BMI and WC were significantly lower than the WE_2 cohort, there were no significant differences in %FM, which further illustrates the inadequacies of BMI, and also suggests that additional measures of WC would not reveal higher levels of %FM

among SAs at equivalent BMI and WC levels when compared to their more contemporary WE_2 counterparts.

The new equation developed in the BIA validation study was applied to a final data set of SA children and adolescents aged 5-14y, to determine absolute (kg) and relative (%) whole body FM and FFM, although application of the correction factor was limited to children aged ≥9y (see section 9.3 Limitations). Sex-specific centile curves were constructed for %FM and %FMM using the original BIA equation and BIA_{reg1} for children aged ≥9y, to make comparisons between the results before and after regression, and also to compare the results with WE_1 and WE_2 (representing high and low incomes respectively) data. Comparisons between SA and high and low income WE datasets were made to ensure that any differences were not simply due to differences in SES. %FFM centile curves using the original BIA regression equation were also constructed for all children aged ≥5y – 14y to make comparisons with the WE_2 dataset across all ages. SMM_a data was used to construct centile curves for a variety of measures.

For both boys and girls overall the WE_1 cohort had the lowest levels of %FM and conversely the highest levels of %FFM than the WE_2 and SA cohort measured at the 50^{th} centile. For boys, use of the original BIA equation indicated that compared to WE_2 children, SA children had very similar levels of %FFM; however, use of BIA_{reg1} revealed that SA boys had less %FFM than WE_2 boys, although very similar levels were observed at age 14y.

For girls, the BIA original equation resulted in more variable results in %FM between WE_1 and SA girls, ranging from SA girls ≤11y having slightly lower levels of %FM than WE_1 girls, to progressively higher %FM at 14y. With BIA_{reg1}, SA girls of all ages had more %FM than both the WE_1 and WE_2. Based on the evidence that BIA underestimates %FM for SAs, and this ethnic group is reported to have higher %FM than WE and other ethnic groups, suggests that the original BIA equation is unsuitable for SAs. However, it is important to note that whilst overall between group (i.e. SA & WE_2) differences increased after application of BIA_{reg1} for boys and girls, this difference was only significant between the male groups. Across specified

age ranges, based on original BIA data, %FFM differences between SA and WE_2 cohorts were only significant between girls in the 11-13y age range, with SA girls having 1.58% less FFM than the WE_2 girls.
Further comparisons between the SA and WE_2 cohort for SMM_a measures (based on original BIA data), revealed that both SA girls and boys had significantly less relative SMMa than the WE_2 sample, for most of the assessed measures including SMMa%, SMMa/FFM%, and SMMaI, with significantly less MFR, although this was not significant across all ages. Sex-specific analyses of sub-group differences revealed some BC differences between Indian, Pakistani, and Bangladeshi children, together with different sub-group outcomes when compared to the WE_2 cohort. Variations in anthropometric and BC measures at different age ranges made it difficult to draw any conclusions on overall relative health risks by sub-group.

Given that the prevalence of type 2 diabetes mellitus (T2DM) is higher in Pakistani and Bangladeshi ethnic sub-groups in the UK, with some studies reporting that Bangladeshi children have a more adverse health profile in relation to T2DM than Indian or Pakistani children, suggests that there may be within sub-group differences in BC, as has been shown in this study. However, these differences between studies suggest that variations in BC within group are more likely to be associated with lifestyle factors, such as diet, physical activity, and SES, as has been reported by others. Although, this simple sub-group categorisation, whilst convenient, may not be indicative of racial/ethnic differences within this very diverse population, given its history. Whilst BC sub-group comparisons led to inconsistent outcomes across different age-ranges, all three sub-groups when compared to their WE_2 counterparts had a lower BMI with no significant differences in %FM, with significantly less relative SMMa in one or more age ranges, and a lower SMMaI across all age ranges for both sexes.

This result illustrates the importance of measuring SMMa, and lends support to the argument that adiposity is not the sole contributor to T2DM or insulin resistance, particularly among SA children within the normal BMI and Wt range. Whilst other studies have also shown that SAs have a higher FM to lean mass (LM; FM: LM) ratio,

to our knowledge, this is the first study to provide evidence on ethnic differences in BC based on appendicular data.

9.2.3 Developing waist circumference percentile charts for South Asian children & youths designed to pass through adult cut-offs – Chapter 6

Sex-specific WC percentile charts for SA children and adolescents were constructed and compared to previously published centile charts representing the WE_1 (high income) child and adolescent population measured in 1988, together with a more contemporary WE_2 (low income) dataset. WC and BMI SDS comparisons were also made with the UK90 growth reference data. Whilst SA children on average had a lower BMI (SDS) compared to the UK90 reference data, WC (SDS) was higher. Similarly, comparisons between SAs and WE_1 data at the 50^{th} centile revealed a higher WC for SAs. However, comparisons between the WE_2 dataset revealed that SA children and adolescents had significantly lower BMI and WC SDS values than their more contemporary WE_2 peers, which has been observed in other studies and is an indication of the secular increase in abdominal FM.

Whilst the BMI and WC SDS values observed in the SA population would suggest that SA children and adolescents had a lower risk of developing obesity related diseases compared to their WE_2 counterparts, research using MRI scans shows that WC is a weaker indicator of visceral fat than total abdominal fat, and visceral fat is regarded as a greater risk factor for developing metabolic syndrome and T2DM. However, use of WC percentile cut-offs based on IDF recommendations as well as cut-offs proposed for Indian adults at age 16y for determining overweight and obesity identified more SAs in this category than similar BMI percentile cut-offs. Thus, using only the BMI UK90 cut-offs for determining overweight and obesity among all children and adolescents in the UK, as is the current NICE recommendation, would exclude a significant proportion of SAs at risk of overweight and obesity that would be identified by using the appropriate WC cut-offs. SA sub-group comparisons revealed that Pakistanis were the only group with no significant differences compared to their WE_2 peers for all measured variables, with significantly higher WC than the other SA ethnic groups. Other studies comparing body composition between SA sub-groups, have similarly reported within-group differences, which as

previously stated is likely to be an indicator of lifestyle rather than genetic differences.

9.2.4 Ethnic differences in leg length and stature between South Asian & white European children & adolescents – Chapter 8

Following the successful development of a field-based SH measure (Chapter 7), ethnic differences in LL, RLL, and stature between SA and WE (UK90 reference data) children and adolescents were examined. LL was determined by subtracting SH from Ht, and SH and LL measures were converted to %sitting-height ratio (SHR) and %leg-length-to-height ratio (LLHR), to assess proportionality of legs and upper body (head and trunk) relative to total Ht. The overall findings of this study were that SA children and adolescents were on average shorter but had relatively longer legs than WEs. More detailed analyses revealed that pre-pubertal SA children were much closer in Ht to the WE reference population, whilst post-pubertal children were on average shorter, suggesting that the adolescent growth spurt is more inhibited within the SA population. Taller stature was positively associated with measures of obesity (BMI and WC), whilst RLL was negatively associated with obesity.

9.3 Limitations

A limitation of the BIA validation study was that the new equation was only valid for children and adolescents aged 9-18y, as the mean age of the study sample was significantly older than the mean age of the large external dataset. Due to the opportunistic nature of recruitment, age was not controlled for, with a very limited number of volunteers in the younger age range. It has been reported that no single BIA equation is likely to be valid throughout childhood and adolescence, which is likely to be attributed to the decline in hydration of FFM during growth and maturation. Children >14y were excluded from the analysis, as both the WE_2 and SA data sets had very few children above this age.

Due to inconsistent results from the ADP, this reference method was excluded. The ADP outcome was attributed to some subjects not willing to adhere to the strict requirement to wear skin-tight clothing. Discrepancies between DXA Wt and scale

Wt were likely to be a further source of error in the validation study, as a correction factor had to be applied to DXA sum of parts, which has been observed in other studies. The 4-C model is considered the 'gold-standard' for in-vivo BC assessment; however, this method requires use of multiple methodologies that are prohibitive in many research settings.

Due to a lack of data for children ≥14y in the large external SA dataset application of the new BIA prediction equation in Chapter 6 and the BC reference centiles that were developed were limited to children aged 9y -14y. The number of children in some of the sub-group comparisons was also very limited, particularly within the Pakistani sub-group, which may have impacted the statistical outcomes. %FM and anthropometric (including BMI) comparisons between WE and SA children were based on SDS from the UK90 reference data, however, as there was no similar reference data available for %FFM or SMMa measures, comparisons were made by age-range which is not as accurate. Large limits of agreement between DXA and BIA observed in this and other studies, indicates that BCA using BIA at an individual level should be treated with caution, and additional methods of assessment should be used before drawing any definitive conclusions on an individual's health status in relation to BC. In certain age ranges there were also limited numbers of participants from certain SA ethnic sub-groups, to allow for any within-group analyses to be made at individual age ranges.

A limitation of the study on stature and LL (Chapter 8), was that the UK90 reference population were measured in 1995-96, and may not now be representative of a contemporary WE population. Based on evidence of secular increases in Ht, differences between the SA and WE populations may have been further enhanced had comparisons been made with a more contemporary WE cohort. Inter-observer differences in measurement of SH between the WE and SA cohorts could not be controlled for, and may also have impacted the findings. RLL data was not available for the UK90 reference data and thus had to be derived from mean LL and stature data, which may have introduced further error. Ethnic sub-group comparisons were not made due to the small sub-group sample sizes.

9.4 Future Research

The ethnic-specific BIA (BC-418MA) prediction equation developed in chapter 4, for the SA child and adolescent population was found only to be valid for children >9y, and as this study and other research shows, no single equation can be used across this whole population group, further studies need to be conducted to assess the validity of prediction equations within narrower age ranges or to assess whether pre- and post-pubertal prediction equations are sufficient. Additional research needs to include sufficient numbers across the whole child and adolescent age range (i.e. 5-18y), as the large external dataset on which the prediction equation was applied had too few children >14y. Improvement in the accuracy of BC using BIA technology is required if it is to be used with confidence as an adjunct to BMI, WC and other field-based methods for individual BCA. Discrepancies between DXA and scale-Wt found in this study as well as others, needs further investigation, if DXA is to be used as a reliable reference method.

Current percentile cut-offs for %FM to define overfat and obesity are derived in part from BMI IOTF cut-offs, rather than clinical evidence. Clinical research would be required to determine appropriate body-fat cut-offs for child and adolescent populations, which may vary by ethnicity due to BC and fat-patterning differences. Given that levels of overweight and obesity are rising, percentile cut-offs may not provide a true indication of health risk associated with actual body fatness.

Further studies assessing SMMa are required to verify the findings in this thesis, particularly as DXA Wt was found to be significantly greater than scale Wt and thus only original BIA data could be used, as limb Wt could not be determined. Improvements in DXA technology are also required to overcome difficulties in marking out limbs for the determination of SMMa, and the fact that large individuals did not fit within the scanning area. Evidence of differences between and within DXA models in BC assessment also requires further investigation, to improve reliability and accuracy when using this reference method.

To verify the findings in chapter 8 on ethnic differences in stature, LL, and RLL, would require comparisons with a more contemporary WE child and adolescent population, particularly as data on RLL was not available. It is also essential that all methods used for BC assessment including anthropometry, are standardised and guidelines closely adhered to, to ensure that the accuracy and reliability of results are not compromised by measurement error.

9.5 Final Overview

The research conducted in this thesis provides some valuable information on SA BC, particularly ethnic differences between the SA and WE child and adolescent population, revealing that compared to a more contemporary WE_2 cohort, SAs at a significantly lower BMI and WC had no significant difference in %FM, but had significantly lower SMM_a. These findings illustrate that BMI is particularly unsuitable for identifying obesity and its associated risks in SAs, and similarly WC offers only a proxy measure for abdominal adiposity. Whilst BIA still has limitations in terms of accuracy in measuring BC at an individual level, recent improvements in BIA tetrapolar devices such as the Tanita BC418-MA, offer advantages in determining BC at a population level that go far beyond BMI.

In the UK, whilst current NICE guidelines advise that BMI centile cut-offs for children and adolescents in ethnic minority groups be used with 'caution', use of WC as an additional measure to assess long-term health risks, is left to the discretion of the health practitioner, with no other BC measures recommended at present. For effective preventative measures of obesity to be introduced it is essential that clear guidelines be developed that enable health practitioners to follow best practice at all times. Whilst WC is a useful additional measure of obesity, BIA provides more comprehensive BC information particularly for the SA population, where both BMI and WC have been shown to be inadequate in this ethnic group.

Further research is required to confirm or refute whether SA children and adolescents have relatively longer legs than their WE counterparts. However, if further studies confirm SAs have a longer RLL, whether SMMa is sufficiently modifiable to offset the health risks associated with the 'thin-fat' phenotype would

need to be investigated. From the results of the studies conducted in this thesis and other supportive research evidence, it would seem prudent for health practitioners to ensure that SA children and adolescents engage in a physically active lifestyle that helps to improve SMM_a.

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Appendices

Appendix A: DXA Scan & Radiation Doses

Appendix A1: DXA Norland_XR_800_Whole_Body_Scanner

(Image copied from PDF - www.inmed.com)

Performance:

Scan Sites:	Whole Body (BMD) with body composition, AP spine, lateral spine hip, and forearm; optional research and small subject.
Scan Time:	5-min. Whole Body, <1.5 minutes AP Spine, <2.0 minutes Hip, <3.0 minutes forearm, <4.0 minutes lateral spine.
Precision:	1.0% - AP spine, 1.2% - hip, 0.8% - forearm, 2.4% - lateral (2 vertebra) whole body BMD-1%.
Accuracy:	1% (based on hydroxyapatite phantom).
Spatial Resolution (Point/ Line):	AP Spine 1.5 mm (1.0 mm selectable) /1.5 mm (1.0 mm selectable), lateral Spine 1.0 mm / 1.0 mm, proximal femur 1.0 mm / 1.0 mm Forearm 1.0 mm / 1.0 mm.
Scan Dose (mRems):	<1.0 mRem typical (AP spine, hip, forearm) and <4 mRem typical (Lateral), <0.1 mRem whole body.
Calibration:	Automatic with supplied 77-Step calibration standard & QC phantom.

Appendix A2: DXA Radiation Doses

(Image copied from DEXA-risk and benefit-assessment (Ward & Knapp, 2007)

Table 1. Radiation doses from DEXA compared to other activities

	Effective dose (µSv)
AP spine*	0.7
Hip*	0.68
Total body*	0.5
Lateral spine*	0.6
Chest x-ray	20
Lateral lumbar spine x-ray	700
Daily background radiation	6 – 20 (depending on location)
Return transatlantic flight	80

*GE Lunar scan doses for a typical patient size using standard mode

Table 2. Activities carrying a risk comparable to receiving an effective dose of 10 µSV from a DEXA scan*

Exposure to natural background radiation for 2 days
Smoking a cigarette
Travelling 30miles by car
Travelling 150miles by aeroplane
Working in a factory for a week

*The Lunar Prodigy protocol of scanning the femur, AP spine and whole body equates to = 1.88µSv. Indeed, the annual risk of death by natural causes (age 40) is 1 in 700, whilst the risk of death from an effective dose of 2µSv is 1 in 10 million.

Appendix B1: Ethical Approval

Ethics and progress review

Julie Hart <Julie.Hart@londonmet.ac.uk>
To: mas1896@my.londonmet.ac.uk

Dear Jabeen

Chris Branford-White has confirmed that you have ethics approval for your project.

We don't yet have a date for the FLS Progress Group but it is likely to take place in December so we will be asking you to submit some work in November.

Best
Julie

--
Julie Hart
Research Office Manager
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http://www.londonmet.ac.uk/research
http://www.londonmet.ac.uk/enterprise
http://www.londonmet.ac.uk/studententerprise
http://www.londonmet.ac.uk/accelerator
http://www.londonmet.ac.uk/metropolitanworks

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London Metropolitan University: Transforming Lives, Meeting Needs, Building Careers

Appendix B2: Example of Flyer

LONDON metropolitan university

Working towards a healthier future

South Asian youths wanted for health research project

- *We are looking for volunteers to take part in a research study to measure body fat and muscle because they are linked with the risk of diabetes and heart disease in South Asians.*
- *To be able to take part you need to be aged 5-22 years, in good health and you (or your parents) need to be originally from India, Pakistan or Bangladesh.*
- *You would need to attend the Science Centre at the university for approximately 1 hour on one occasion only.*
- *You will find out how much muscle and fat you have in your body and where this is located, as well as contributing to important health research.*
- *All reasonable travel costs will be refunded and in appreciation of your time, you will receive a JD Sports voucher worth £10.*
- *The university Science Centre is located at: 29 Hornsey Road, London N7 7DD.*
- *If you are interested in taking part please complete and return the tear-off reply slip below or contact **Jabeen Shah on 07425-150419** and you will then be provided with a full information pack and consent form.*

The main investigator is: Jabeen Shah BSc (Hons) MSc and the research will be conducted under the direction of Dr. David McCarthy (Professor of Nutrition & Health).

This research has been reviewed and ethical clearance has been granted by the Research Ethics Office at London Metropolitan University.

.....cut along dotted line.....

South Asian Health Research Project at London Metropolitan University

Name (print full name).....

Address:.....

Tel:.....email.....

Please return reply slip to: Dr David McCarthy, London Metropolitan University, Tower Building, TB9/2, 166-220 Holloway Road, London N7 8DB

Appendix B3: Sample Letter of Invitation

London Metropolitan University
London North Campus
166-220 Holloway Road
London N7 8DB

Date: 2011 **Letter of invitation**

Dear All,

I am a health researcher at London Metropolitan University, and am writing to request permission for you (if aged 16-22 years) or your child (if over 5 years) to take part in an important South Asian health study. South Asians (people originally from Pakistan, India and Bangladesh) have a much higher risk of developing type 2 diabetes and heart disease than white European people. **Our children are 13 times at greater risk of getting type 2 diabetes than white European children, and we are 50% more likely to die of heart disease than white European people** (see attached). The underlying causes develop slowly during childhood. My research is to try and understand what physical differences there are between South Asian and white European children and youths, to try and identify what can be done to reduce the risk of getting type 2 diabetes and heart disease, with the aim of protecting and improving the health and well being of future generations.

South Asian volunteers aged 5-22 years are required for my study which has received full ethical approval from London Metropolitan University. This will only involve taking some simple physical measurements including weight, height and waist, and also involve standing barefoot on a special set of scales to measure body muscle and fat. To test the accuracy of the scales used to measure body muscle and fat, some participants, will also be asked to take part in a further very quick and simple measurement using two more advanced methods - the BodPod, and the DXA scanner (full details attached). All the methods used are safe and there is no discomfort to participants. BodPod and DXA measurements will take place at the university Science Centre located on Hornsey Road (N77DD); volunteers for this part of the study will have all transport costs refunded and will also receive a £10 JD Sports voucher in appreciation of their time. Taking part in this study is entirely voluntary and all measurements will be taken in strict confidence with a parent/carer present for children. You/your child may withdraw from all/part of the study at any time at his/her/your request by contacting Jabeen Shah (contact details attached) and detailing the ID number on your form.

If you and/or your child are happy to participate, please complete the attached consent form and return it to your participating centre or contact Jabeen Shah by telephone. We will provide any feedback you require on your/your child's measurements. Thank you for taking the time to read this letter, and I hope you will agree to participate in this important health research project.

Yours sincerely,

Jabeen Shah BSc (Hons) MSc
Obesity Researcher

Dr. H. David McCarthy
Professor of Nutrition & Health

Please note if you have any concerns or questions about this project please contact Jabeen Shah. However, if you would like to raise any concerns with an independent party please contact the project supervisor (contact details overleaf).

Investigator contact details:

Name: Jabeen Shah

Address: Institute for Health Research & Policy, London Metropolitan University, Tower Building, TB9/2, 166-220 Holloway Road, London N7 8DB

Email:mas1896@my.londonmet.ac.uk

Telephone: 07425-150419

Project supervisor contact details:

Name: Professor David McCarthy

Address: Institute for Health Research & Policy, London Metropolitan University, Tower Building, TB9/2, 166-220 Holloway Road, London N7 8DB

Email: d.mccarthy@londonmet.ac.uk

Telephone: 020 7133 2547

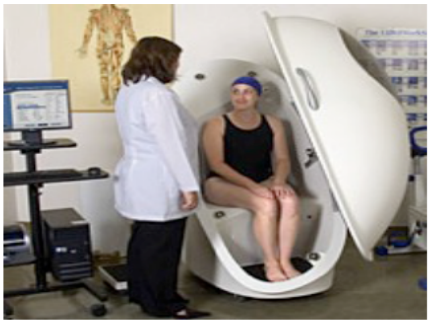
Appendix B4: Background Information Sheet on BIA, ADP, and DXA

London Metropolitan University - Background information on subject tests for South Asian Health Research Study 2011

- **Bioelectrical Impedance Scales**

A Bioelectric Impedance Analyzer (BIA) is a sophisticated scientific instrument, used in research and analysis, which measures the resistance of body tissues to the flow of a small harmless electrical signal. The proportion of body fat can be calculated as the current flows more easily through the parts of the body that are composed mostly of water (such as blood, urine & muscle) than it does through bone, fat or air. It is possible to predict how much body fat a person has by combining the bioelectric impedance measure with other factors such as height, weight, gender, fitness level and age. On a budget level, many bathroom type weighing scales are also available with a BIA, which is two built-in footpad electrodes on the base of the scale which the person stands on.

- **The Bod Pod**



The BOD POD is a computerized, egg-shaped chamber which measures a subject's mass and volume, from which their whole-body density is determined. Using these data, body fat and lean muscle mass can then be calculated. You will be required to wear a swim suit or full body skin-tight suit and swim cap. A few swim suits and swim caps are available.

All measurements are taken in a private room and a gown is provided when getting in and out of the chamber. A female or male researcher is available to conduct the test in full privacy, as required by the participant. The test should take no longer than 10 minutes.

For more information go to: http://www.bodpod.com/products/howWorkBodpod

London Metropolitan University - Background information on subject tests for South Asian Health Research Study 2011

- **The DXA Scan**

A dual energy X-ray (DXA) scan is a type of X-ray that measures the amount of calcium in bones. It is also used to measure the relative amounts of body fat and muscle. It uses a much lower level of radiation, equivalent to less than one day's exposure to natural background radiation, and is considered safe, although not recommended during pregnancy.
A DXA scan is quick and painless, usually lasting no longer than 10 minutes. Tests will be done in subject's own clothing.

For more information go to: http://www.cks.nhs.uk/patient_information_leaflet/dxa_scan/introduction

Appendix B5: Participant Consent Form

ID number_____

PARTICIPANT CONSENT FORM

By completing and signing this consent form you are agreeing to the following points:

- I have read and received a copy of the details of this research project, and have been given the opportunity to ask questions.
- I understand that after consenting to my child taking part in this study you will provide a full explanation of the procedures to be followed, and will answer any questions I may ask.
- I understand that my child has the right to withdraw from the study at any point at my and/or his/her request for any reason.
- All measurements will be taken in the strictest confidence.
- I understand that my child's identity will be protected and not be identified in any published results.
- I would also be happy to consider my child taking part in the BodPod and/or DXA test once I have understood and accepted details of what is involved. (Please tick if yes or cross if no in box) ☐

Child's name.....

Signature of parent/guardian (if under 16y) or participant (if aged16y or over)

.....

Contact
address:.....
.....
.....

Contact telephone
number(s):.....
.....

Date.....

Please note if you have any concerns or questions about this project please contact Jabeen Shah. However, if you would like to raise any concerns with an independent party please contact the project supervisor.

Appendix C: SPSS Outputs Showing Tests of Normality

Tests of Normality[a] SA Boys - Validation Study

	Kolmogorov-Smirnov[b]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
weight z score	.149	27	.128	.965	27	.468
BMI z score	.113	27	.200*	.982	27	.900
Waist circumference (cm) Z score	.136	21	.200*	.951	21	.350
BIA % body fat z score(UK90)	.158	25	.107	.956	25	.348
Height (m) z score	.091	27	.200*	.964	27	.447

*. This is a lower bound of the true significance.

a. Gender = male; b. Lilliefors Significance Correction

Tests of Normality[a] SA Girls – Validation Study

	Kolmogorov-Smirnov[b]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
weight z score	.097	24	.200*	.946	24	.217
BMI z score	.116	24	.200*	.950	24	.276
Waist circumference (cm) Z score	.129	19	.200*	.946	19	.337
BIA % body fat z score (UK90)	.113	23	.200*	.970	23	.698
Height (m) z score	.102	24	.200*	.979	24	.868

*. This is a lower bound of the true significance.

a. Gender = female; b. Lilliefors Significance Correction

Tests of Normality[a] – SA external dataset (Boys)

	Kolmogorov-Smirnov[b]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
Z-score height	.020	691	.200*	.996	691	.110
Z-score weight	.028	691	.200*	.996	691	.088
Z-score BMI	.046	691	.002	.992	691	.001
Z-score waist circimference	.055	683	.000	.989	683	.000
Z-score % body fat	.039	681	.017	.987	681	.000

*. This is a lower bound of the true significance.

a. Gender = male, SA or WE ethnic group = South Asian; b. Lilliefors Significance Correction

Tests of Normality[a] WE external dataset (Boys)

	Kolmogorov-Smirnov[b]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
Z-score height	.027	598	.200*	.995	598	.076
Z-score weight	.032	598	.200*	.994	598	.017
Z-score BMI	.047	598	.003	.993	598	.005
Z-score waist circimference	.054	586	.000	.985	586	.000
Z-score % body fat	.029	589	.200*	.985	589	.000

*. This is a lower bound of the true significance.

a. Gender = male, SA or WE ethnic group = White European

b. Lilliefors Significance Correction

Tests of Normality[a] SA external dataset (Girls)

	Kolmogorov-Smirnov[b]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
Z-score height	.021	933	.200*	.998	933	.281
Z-score weight	.020	933	.200*	.998	933	.589
Z-score BMI	.024	933	.200*	.997	933	.041
Z-score waist circimference	.029	926	.060	.997	926	.043
Z-score % body fat	.026	921	.139	.998	921	.276

*. This is a lower bound of the true significance.

a. Gender = female, SA or WE ethnic group = South Asian

b. Lilliefors Significance Correction

Tests of Normality[a] WE external dataset (Girls)

	Kolmogorov-Smirnov[b]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
Z-score height	.041	543	.031	.994	543	.040
Z-score weight	.039	543	.044	.994	543	.022
Z-score BMI	.050	543	.003	.990	543	.001
Z-score waist circimference	.046	532	.009	.990	532	.001
Z-score % body fat	.043	532	.019	.996	532	.143

a. Gender = female, SA or WE ethnic group = White European

b. Lilliefors Significance Correction

Appendix D: SPSS Outputs Showing Tests of Normality for Ht, SH, & LL

Tests of Normality[a] SA Boys

	Kolmogorov-Smirnov[b]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
SDS - Height	.065	108	.200*	.989	108	.514
SDS - Sitting Height	.068	108	.200*	.990	108	.639
SDS - Leg length	.048	108	.200*	.988	108	.461

*. This is a lower bound of the true significance.

a. Gender = Male

b. Lilliefors Significance Correction

Tests of Normality[a] SA Girls

	Kolmogorov-Smirnov[b]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
SDS - Height	.061	63	.200*	.989	63	.837
SDS - Sitting Height	.050	63	.200*	.989	63	.850
SDS - Leg length	.093	63	.200*	.980	63	.386

*. This is a lower bound of the true significance.

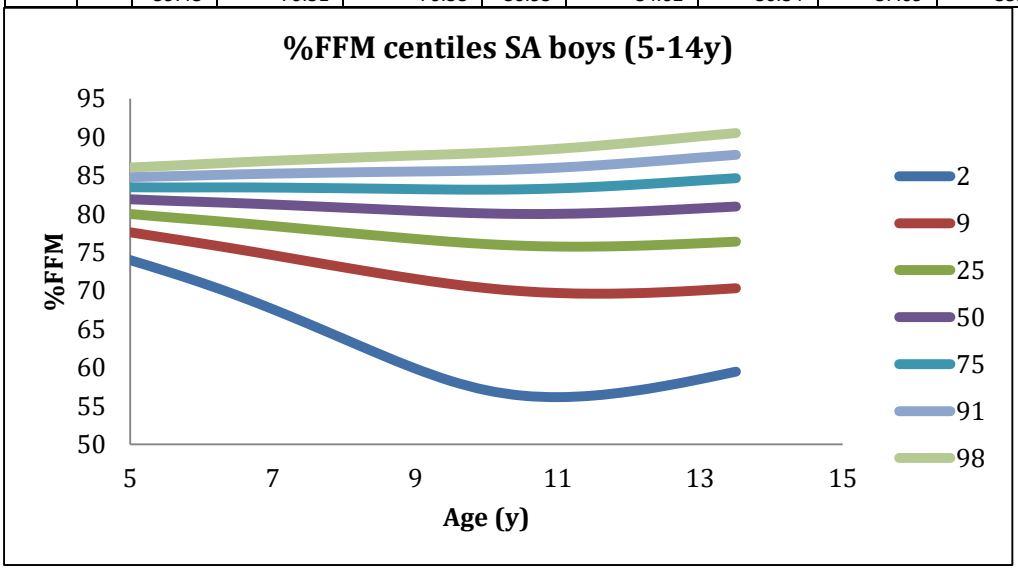
a. Gender = Female

b. Lilliefors Significance Correction

Appendix E: Ethnic Differences in Body Composition

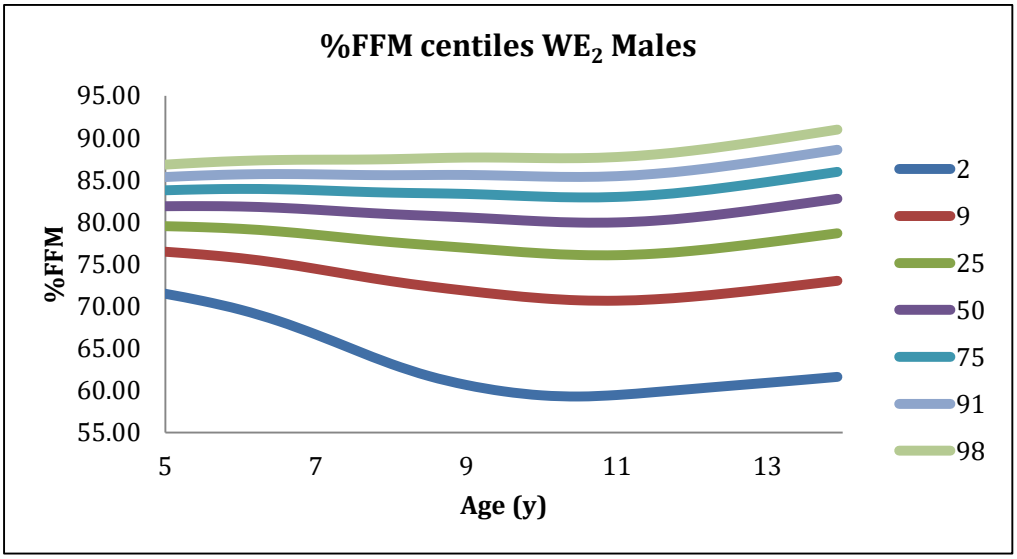
%FFM Centiles SA Boys

Age	n	-2.05	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
		2	9	25	50	75	85	91	95	98
4.5	21	75.06	78.18	80.31	82.05	83.49	84.18	84.72	85.23	85.88
5	45	73.97	77.59	79.98	81.89	83.46	84.20	84.79	85.33	86.03
5.5	30	72.55	76.87	79.59	81.71	83.44	84.25	84.88	85.48	86.23
6	41	71.04	76.14	79.21	81.55	83.44	84.32	85.01	85.65	86.46
6.5	49	69.39	75.40	78.83	81.39	83.44	84.39	85.13	85.82	86.69
7	48	67.60	74.62	78.42	81.21	83.42	84.44	85.23	85.97	86.89
7.5	35	65.69	73.81	77.99	81.01	83.38	84.47	85.31	86.10	87.08
8	34	63.72	73.02	77.56	80.80	83.33	84.49	85.39	86.22	87.26
8.5	38	61.75	72.25	77.14	80.60	83.28	84.51	85.45	86.34	87.44
9	39	59.88	71.52	76.74	80.39	83.22	84.51	85.51	86.44	87.59
9.5	29	58.26	70.86	76.36	80.20	83.17	84.52	85.57	86.54	87.75
10	46	57.06	70.32	76.06	80.06	83.15	84.57	85.66	86.67	87.94
10.5	38	56.36	69.93	75.85	79.99	83.20	84.67	85.80	86.86	88.17
11	32	56.17	69.69	75.74	80.00	83.31	84.84	86.01	87.11	88.47
11.5	35	56.37	69.60	75.74	80.10	83.50	85.07	86.28	87.41	88.82
12	19	56.89	69.64	75.82	80.26	83.74	85.36	86.60	87.77	89.22
12.5	31	57.62	69.80	75.97	80.47	84.03	85.68	86.96	88.16	89.66
13	42	58.51	70.03	76.17	80.71	84.33	86.02	87.33	88.56	90.10
13.5	13	59.48	70.31	76.38	80.95	84.62	86.34	87.69	88.94	90.52



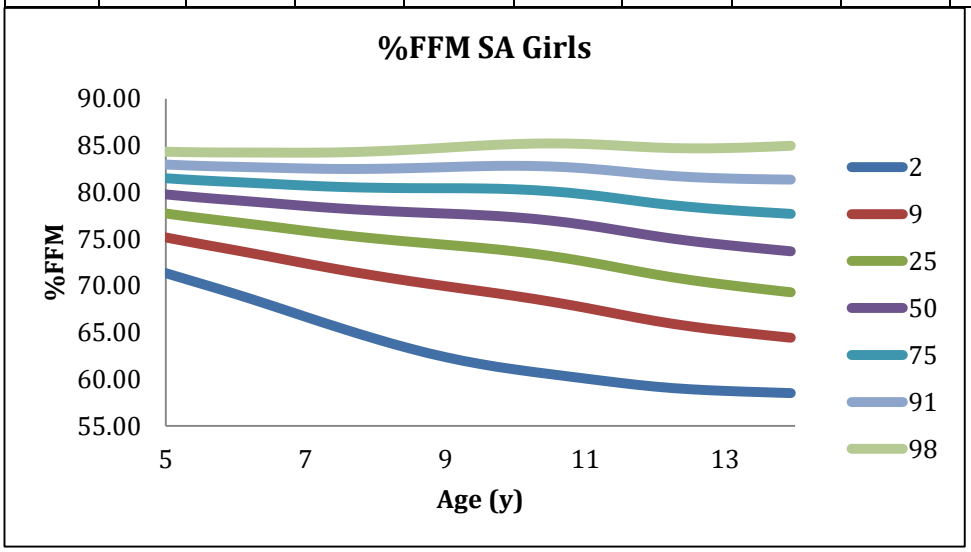
%FFM centiles WE Boys

Age	N	-2.053749	-1.340755	-0.674897	0	0.674897	1.036433	1.340755	1.644854	2.053749
		2	9	25	50	75	85	91	95	98
4.5	24	72.27	76.78	79.62	81.83	83.64	84.49	85.15	85.77	86.55
5	19	71.49	76.47	79.53	81.87	83.77	84.66	85.36	86.01	86.83
5.5	43	70.63	76.13	79.40	81.89	83.88	84.82	85.54	86.22	87.08
6	43	69.59	75.71	79.21	81.84	83.93	84.91	85.67	86.38	87.26
6.5	43	68.27	75.14	78.91	81.69	83.90	84.92	85.71	86.45	87.38
7	33	66.66	74.44	78.50	81.45	83.78	84.85	85.68	86.45	87.41
7.5	40	64.89	73.69	78.05	81.18	83.62	84.74	85.61	86.42	87.42
8	41	63.17	72.97	77.63	80.93	83.49	84.67	85.58	86.42	87.47
8.5	37	61.73	72.36	77.27	80.74	83.42	84.65	85.60	86.48	87.58
9	28	60.67	71.83	76.94	80.55	83.34	84.62	85.60	86.52	87.67
9.5	33	59.91	71.34	76.60	80.31	83.19	84.51	85.53	86.48	87.66
10	30	59.44	70.94	76.29	80.07	83.02	84.37	85.42	86.39	87.60
10.5	37	59.31	70.69	76.09	79.93	82.93	84.30	85.37	86.36	87.60
11	31	59.48	70.65	76.07	79.95	82.98	84.38	85.46	86.47	87.73
11.5	36	59.82	70.81	76.24	80.15	83.22	84.64	85.73	86.76	88.03
12	36	60.20	71.13	76.59	80.53	83.63	85.06	86.17	87.20	88.49
12.5	19	60.57	71.56	77.06	81.03	84.16	85.60	86.72	87.77	89.07
13	18	60.93	72.04	77.60	81.61	84.76	86.22	87.35	88.40	89.72
13.5	3	61.30	72.56	78.17	82.22	85.40	86.87	88.01	89.07	90.40



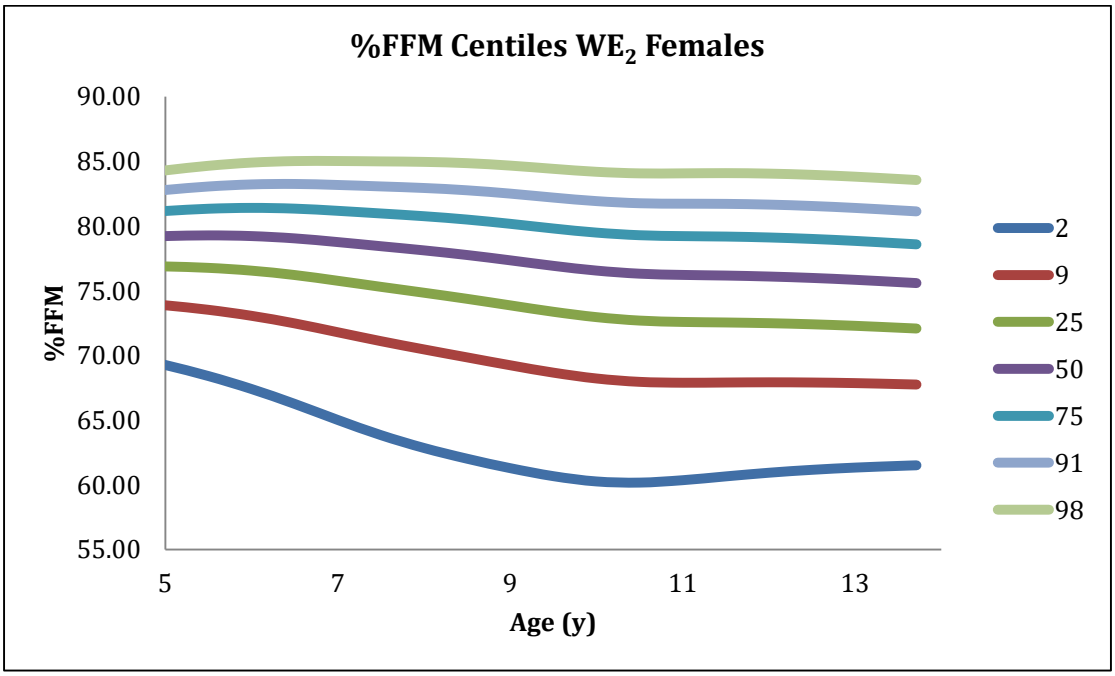
%FFM centiles SA Girls

Age	N	2	9	25	50	75	85	91	95	98
4.5	21	72.47	75.86	78.21	80.13	81.74	82.51	83.12	83.69	84.41
5	27	71.35	75.15	77.71	79.77	81.49	82.31	82.96	83.56	84.33
5.5	40	70.24	74.45	77.23	79.44	81.27	82.13	82.81	83.45	84.26
6	37	69.11	73.78	76.78	79.14	81.08	82.00	82.72	83.39	84.24
6.5	32	67.92	73.08	76.32	78.84	80.90	81.87	82.63	83.34	84.23
7	55	66.68	72.37	75.85	78.54	80.71	81.74	82.53	83.28	84.22
7.5	34	65.47	71.69	75.42	78.26	80.57	81.64	82.48	83.27	84.26
8	37	64.33	71.06	75.03	78.05	80.48	81.62	82.50	83.33	84.37
8.5	37	63.27	70.48	74.69	77.87	80.44	81.64	82.57	83.45	84.55
9	31	62.37	69.94	74.37	77.72	80.43	81.69	82.68	83.60	84.77
9.5	38	61.63	69.43	74.04	77.56	80.40	81.74	82.78	83.76	84.99
10	35	61.04	68.89	73.65	77.32	80.31	81.72	82.82	83.86	85.16
10.5	30	60.54	68.30	73.17	76.97	80.11	81.59	82.75	83.85	85.23
11	104	60.07	67.64	72.57	76.50	79.76	81.32	82.55	83.70	85.16
11.5	104	59.61	66.93	71.88	75.91	79.30	80.93	82.22	83.44	84.98
12	87	59.21	66.24	71.19	75.31	78.82	80.52	81.87	83.15	84.78
12.5	103	58.92	65.66	70.60	74.79	78.43	80.21	81.62	82.97	84.70
13	49	58.75	65.19	70.10	74.37	78.13	79.98	81.47	82.89	84.72
13.5	14	58.61	64.78	69.66	73.99	77.89	79.83	81.39	82.89	84.84



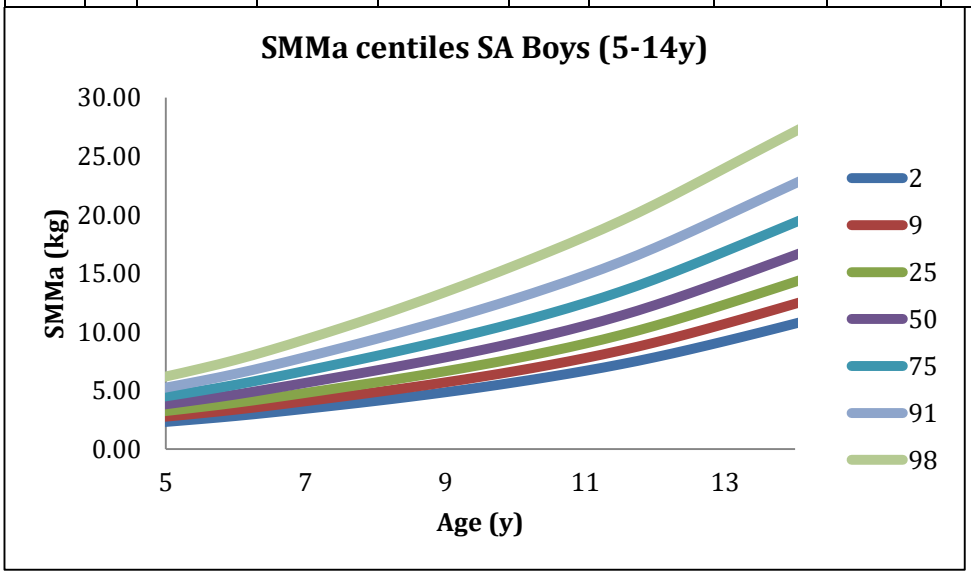
%FFM centiles WE_2 Girls

Age	N	-2.05	-1.34	-0.67	0	0.67	1.04	1.34	1.64	2.05
		2	9	25	50	75	85	91	95	98
4.51	22	70.04	74.20	76.95	79.15	80.96	81.82	82.50	83.14	83.94
5	23	69.28	73.90	76.88	79.23	81.16	82.08	82.79	83.46	84.31
5.5	35	68.42	73.54	76.77	79.28	81.34	82.30	83.06	83.77	84.66
6	41	67.42	73.08	76.55	79.23	81.40	82.42	83.22	83.97	84.91
6.5	29	66.27	72.49	76.21	79.05	81.34	82.42	83.26	84.04	85.02
7	42	65.02	71.81	75.77	78.77	81.17	82.30	83.18	84.00	85.03
7.5	28	63.85	71.12	75.30	78.45	80.97	82.15	83.07	83.92	85.00
8	34	62.85	70.48	74.84	78.13	80.76	81.99	82.94	83.84	84.96
8.5	32	62.02	69.87	74.37	77.77	80.50	81.77	82.77	83.70	84.86
9	35	61.29	69.26	73.87	77.36	80.17	81.49	82.51	83.47	84.68
9.5	32	60.67	68.69	73.37	76.93	79.81	81.15	82.21	83.19	84.43
10	31	60.26	68.24	72.96	76.56	79.49	80.86	81.93	82.94	84.20
10.5	39	60.18	67.98	72.69	76.33	79.29	80.68	81.77	82.79	84.08
11	35	60.36	67.91	72.59	76.23	79.22	80.63	81.73	82.77	84.08
11.5	24	60.66	67.93	72.55	76.18	79.18	80.61	81.72	82.77	84.10
12	20	60.94	67.94	72.49	76.11	79.11	80.54	81.66	82.72	84.06
12.5	22	61.16	67.93	72.40	76.00	79.00	80.43	81.56	82.62	83.97
13	14	61.34	67.88	72.29	75.85	78.85	80.28	81.41	82.48	83.83
13.5	3	61.47	67.80	72.14	75.68	78.66	80.09	81.22	82.29	83.65
13.71	0	61.51	67.77	72.08	75.60	78.58	80.01	81.14	82.21	83.57



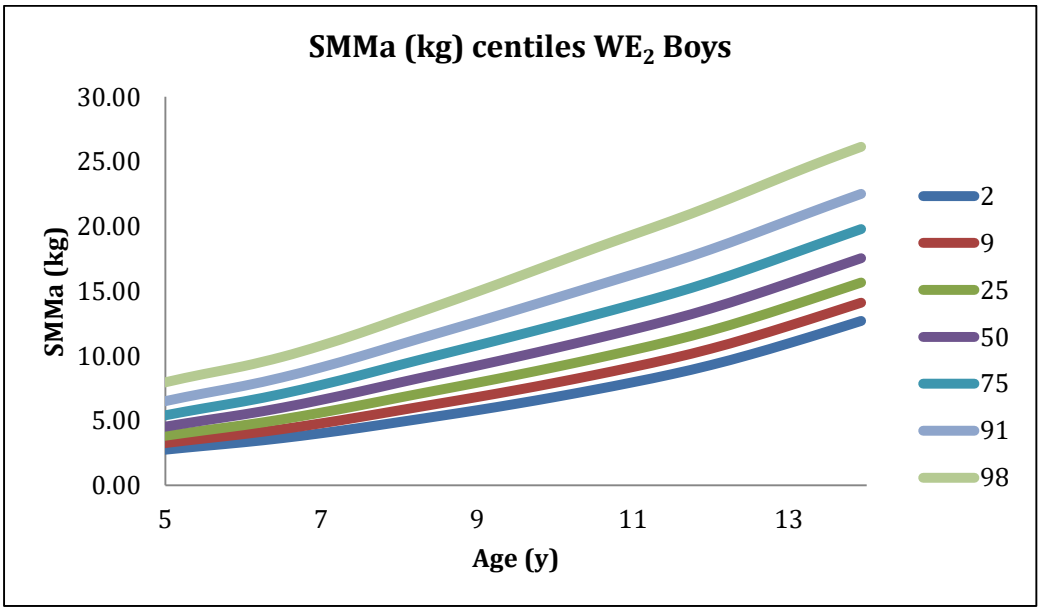
SMMa (kg) centiles SA Boys

Age	N	-2.053749	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
		2	9	25	50	75	85	91	95	98
4.29		1.97	2.36	2.78	3.27	3.83	4.16	4.46	4.78	5.24
4.5	20	2.07	2.48	2.92	3.43	4.02	4.37	4.69	5.03	5.51
5	38	2.31	2.76	3.25	3.83	4.49	4.89	5.24	5.62	6.18
5.5	27	2.55	3.05	3.59	4.23	4.97	5.41	5.81	6.24	6.86
6	38	2.81	3.36	3.96	4.66	5.48	5.98	6.42	6.90	7.60
6.5	41	3.11	3.71	4.36	5.14	6.06	6.61	7.11	7.65	8.44
7	39	3.43	4.08	4.80	5.66	6.67	7.29	7.85	8.46	9.35
7.5	26	3.76	4.46	5.24	6.18	7.29	7.98	8.61	9.29	10.30
8	26	4.10	4.85	5.69	6.71	7.93	8.69	9.39	10.15	11.28
8.5	32	4.46	5.26	6.16	7.26	8.59	9.42	10.20	11.04	12.32
9	37	4.84	5.69	6.65	7.83	9.28	10.19	11.04	11.97	13.39
9.5	23	5.26	6.15	7.18	8.44	10.00	10.99	11.92	12.95	14.52
10	34	5.70	6.65	7.74	9.09	10.77	11.83	12.84	13.96	15.68
10.5	29	6.17	7.19	8.35	9.79	11.59	12.73	13.82	15.02	16.88
11	22	6.68	7.76	9.00	10.55	12.46	13.69	14.85	16.14	18.13
11.5	22	7.23	8.40	9.73	11.38	13.43	14.74	15.97	17.34	19.46
12	18	7.84	9.10	10.53	12.30	14.49	15.89	17.20	18.66	20.89
12.5	30	8.51	9.87	11.41	13.32	15.66	17.14	18.53	20.07	22.42
13	42	9.22	10.69	12.35	14.38	16.87	18.44	19.91	21.53	24.00
13.5	13	9.96	11.53	13.30	15.47	18.11	19.76	21.30	23.00	25.57
14	5	10.72	12.40	14.28	16.57	19.35	21.08	22.69	24.46	27.13
14.5	3	11.49	13.27	15.26	17.67	20.58	22.39	24.06	25.89	28.65
15	4	12.28	14.16	16.25	18.77	21.80	23.67	25.40	27.30	30.12
15.5	4	13.07	15.05	17.24	19.87	23.01	24.94	26.73	28.67	31.57
15.95	3	13.80	15.86	18.13	20.85	24.09	26.08	27.91	29.90	32.85



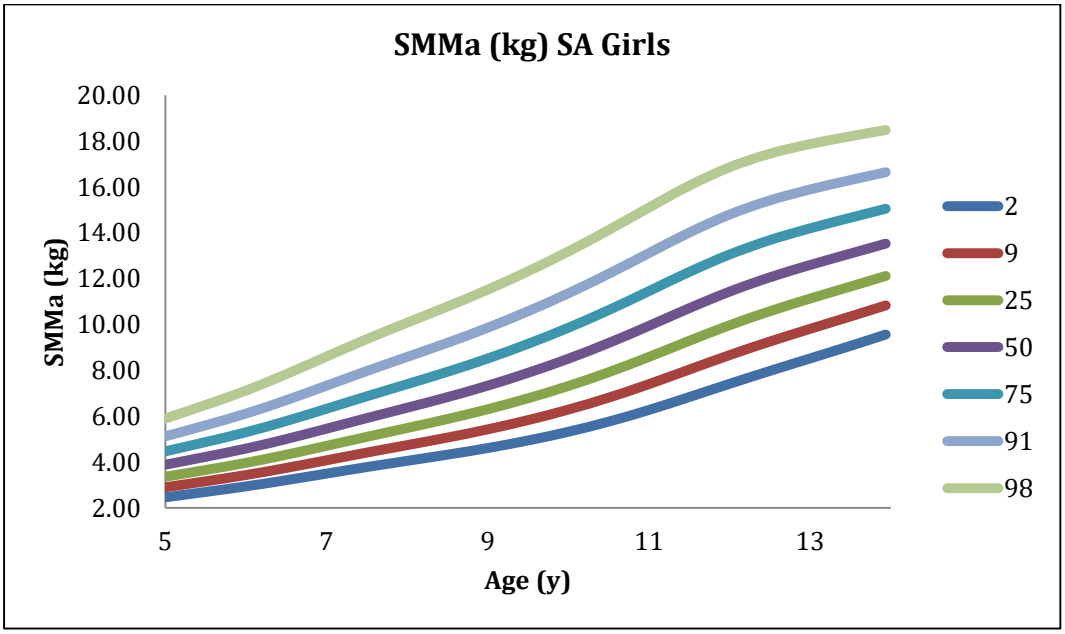
SMMa (kg) centiles WE Boys

Age	N	-2.05	-1.34	-0.67	0	0.67	1.03	1.34	1.64	2.05
		2	9	25	50	75	85	91	95	98
4.47		2.42	2.85	3.35	3.98	4.77	5.28	5.76	6.31	7.16
4.5	24	2.44	2.88	3.38	4.01	4.81	5.32	5.81	6.36	7.21
5	21	2.76	3.26	3.83	4.54	5.41	5.97	6.49	7.07	7.96
5.5	43	3.05	3.61	4.25	5.02	5.96	6.55	7.10	7.70	8.60
6	42	3.33	3.95	4.64	5.48	6.48	7.09	7.66	8.28	9.20
6.5	43	3.66	4.34	5.10	6.00	7.07	7.73	8.33	8.97	9.92
7	33	4.03	4.79	5.62	6.60	7.76	8.46	9.10	9.79	10.79
7.5	40	4.45	5.27	6.18	7.25	8.50	9.26	9.95	10.69	11.76
8	41	4.89	5.79	6.77	7.93	9.29	10.11	10.86	11.66	12.82
8.5	37	5.35	6.30	7.35	8.59	10.05	10.94	11.74	12.61	13.88
9	28	5.81	6.81	7.92	9.24	10.80	11.76	12.63	13.57	14.95
9.5	33	6.30	7.35	8.51	9.91	11.57	12.59	13.53	14.55	16.05
10	30	6.83	7.92	9.13	10.59	12.35	13.44	14.44	15.54	17.18
10.5	37	7.38	8.51	9.77	11.30	13.15	14.30	15.36	16.53	18.29
11	31	7.97	9.14	10.45	12.04	13.96	15.16	16.28	17.51	19.36
11.5	36	8.59	9.80	11.16	12.80	14.80	16.05	17.21	18.49	20.43
12	36	9.29	10.55	11.95	13.66	15.72	17.01	18.22	19.55	21.56
12.5	20	10.10	11.40	12.85	14.60	16.74	18.07	19.32	20.69	22.77
13	18	10.97	12.32	13.81	15.62	17.80	19.17	20.45	21.86	24.00
13.5	3	11.89	13.27	14.80	16.65	18.88	20.28	21.58	23.01	25.18
13.93	0	12.70	14.11	15.67	17.54	19.80	21.20	22.52	23.96	26.15



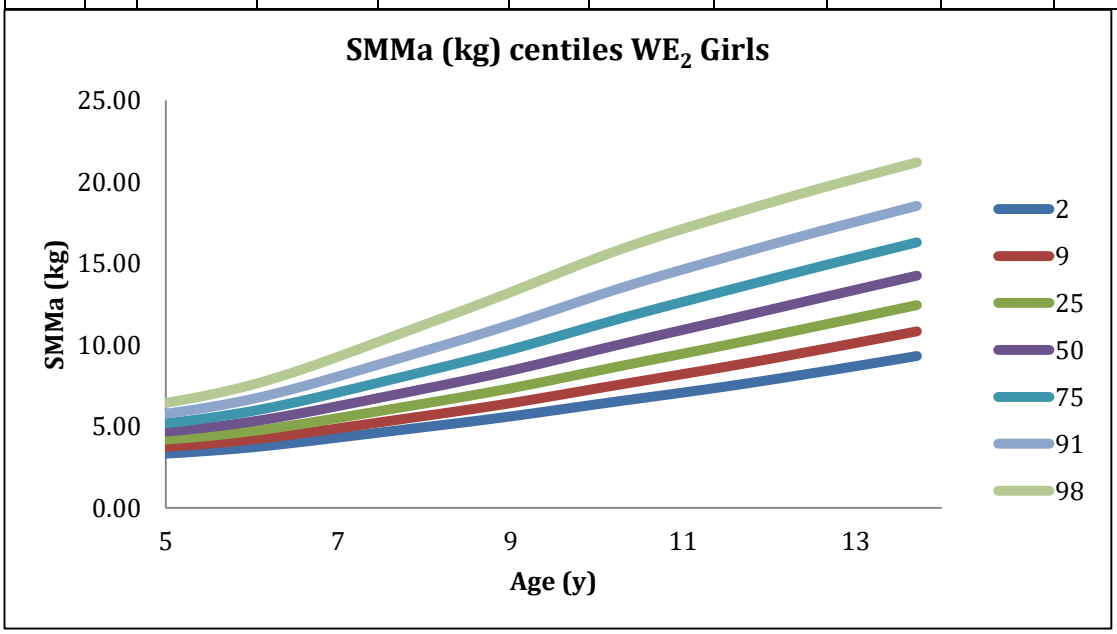
SMMa (kg) centiles SA Girls

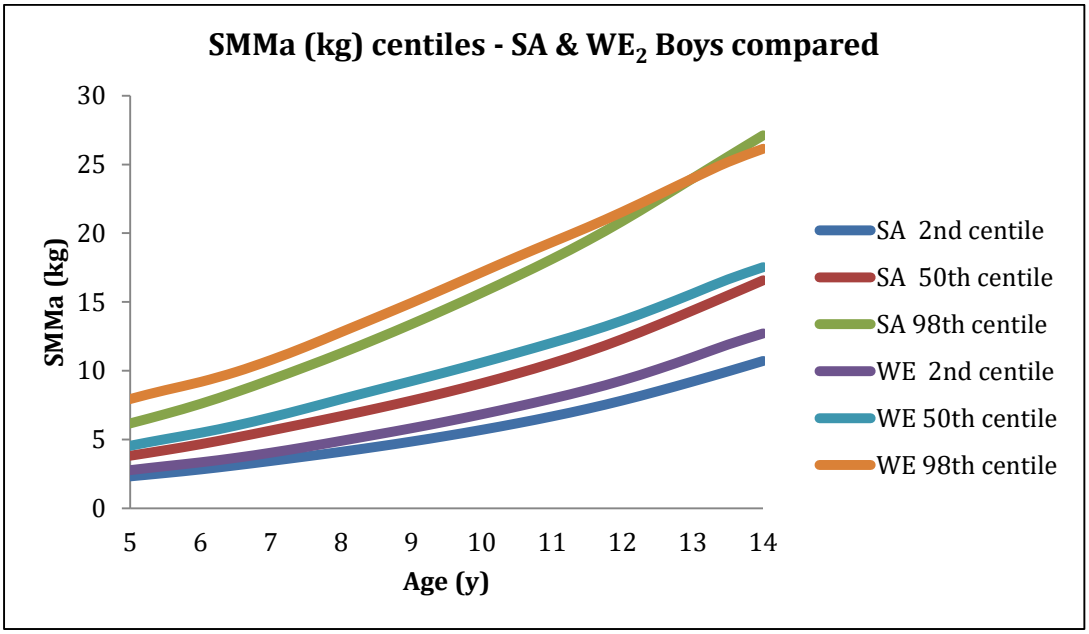
Age	N	-2.05	-1.34	-0.67	0	0.67	1.04	1.34	1.64	2.05
		2	9	25	50	75	85	91	95	98
4.46		2.20	2.60	3.02	3.49	4.02	4.32	4.59	4.87	5.26
4.5	23	2.22	2.62	3.05	3.52	4.05	4.36	4.63	4.91	5.31
5	22	2.46	2.89	3.35	3.88	4.46	4.81	5.12	5.44	5.90
5.5	34	2.69	3.15	3.65	4.22	4.87	5.25	5.60	5.96	6.49
6	29	2.93	3.43	3.96	4.58	5.30	5.72	6.11	6.52	7.11
6.5	24	3.20	3.74	4.31	4.99	5.78	6.26	6.69	7.16	7.83
7	45	3.49	4.07	4.70	5.45	6.32	6.85	7.33	7.84	8.60
7.5	27	3.78	4.41	5.09	5.91	6.86	7.44	7.97	8.53	9.36
8	33	4.05	4.73	5.48	6.36	7.39	8.02	8.58	9.19	10.08
8.5	27	4.32	5.06	5.87	6.82	7.93	8.60	9.20	9.84	10.78
9	19	4.61	5.42	6.30	7.33	8.51	9.22	9.86	10.53	11.52
9.5	28	4.94	5.82	6.78	7.88	9.15	9.90	10.58	11.29	12.32
10	29	5.32	6.28	7.31	8.50	9.85	10.65	11.36	12.11	13.18
10.5	23	5.76	6.80	7.91	9.19	10.62	11.46	12.21	12.99	14.12
11	98	6.27	7.39	8.58	9.93	11.45	12.33	13.11	13.93	15.10
11.5	98	6.83	8.03	9.28	10.70	12.28	13.20	14.01	14.85	16.05
12	87	7.41	8.66	9.96	11.43	13.04	13.97	14.79	15.65	16.86
12.5	106	7.97	9.25	10.58	12.05	13.67	14.60	15.41	16.26	17.45
13	49	8.52	9.81	11.13	12.59	14.18	15.09	15.89	16.72	17.87
13.5	11	9.06	10.35	11.65	13.09	14.64	15.53	16.30	17.10	18.21
13.94	3	9.56	10.83	12.12	13.53	15.04	15.90	16.64	17.41	18.48

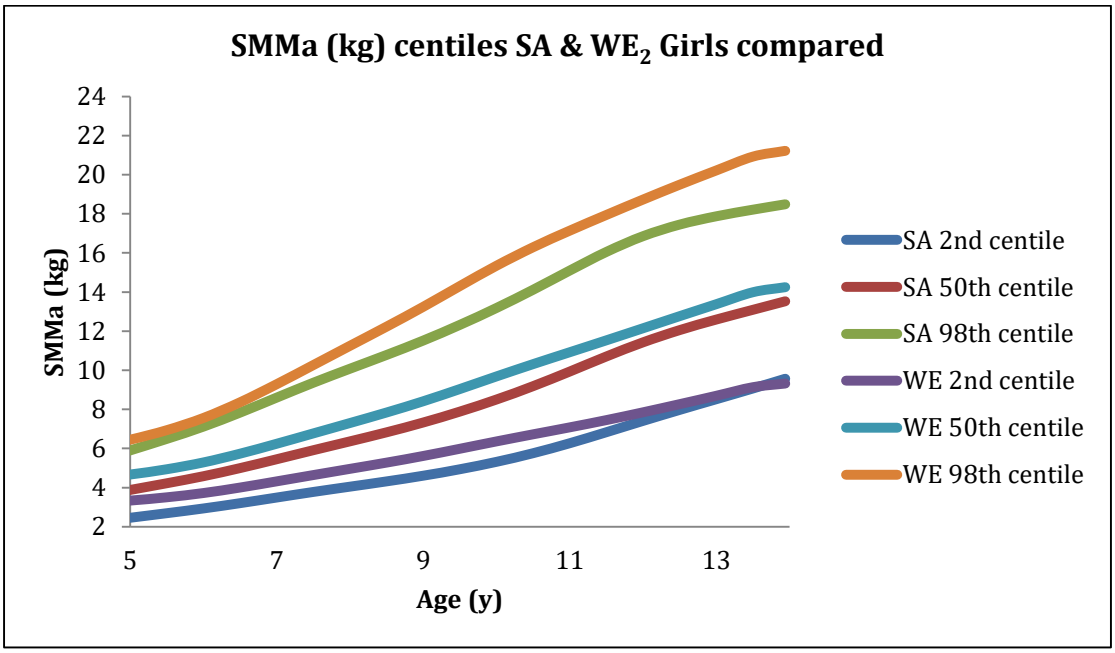


SMMa (kg) centiles WE Girls

Age	N	-2.053749	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
		2	9	25	50	75	85	91	95	98
4.51	22	3.20	3.59	4.00	4.45	4.93	5.21	5.45	5.70	6.05
5	23	3.33	3.75	4.19	4.67	5.20	5.51	5.78	6.06	6.45
5.5	34	3.50	3.95	4.42	4.94	5.53	5.87	6.17	6.49	6.94
6	41	3.72	4.20	4.71	5.29	5.95	6.33	6.68	7.04	7.56
6.5	29	4.00	4.53	5.09	5.74	6.48	6.92	7.32	7.75	8.36
7	42	4.32	4.89	5.52	6.25	7.09	7.60	8.07	8.56	9.29
7.5	28	4.64	5.27	5.96	6.77	7.73	8.31	8.84	9.42	10.27
8	34	4.96	5.65	6.40	7.31	8.37	9.03	9.63	10.28	11.25
8.5	32	5.28	6.03	6.85	7.84	9.02	9.75	10.42	11.15	12.23
9	35	5.62	6.43	7.34	8.42	9.71	10.51	11.25	12.05	13.24
9.5	32	5.98	6.87	7.86	9.04	10.46	11.33	12.13	13.00	14.30
10	31	6.36	7.33	8.41	9.69	11.22	12.16	13.02	13.96	15.34
10.5	39	6.72	7.78	8.94	10.32	11.95	12.94	13.85	14.84	16.29
11	35	7.08	8.22	9.46	10.93	12.65	13.69	14.63	15.65	17.14
11.5	24	7.45	8.67	9.99	11.54	13.33	14.41	15.38	16.42	17.94
12	20	7.85	9.14	10.53	12.15	14.02	15.13	16.13	17.19	18.73
12.5	22	8.27	9.63	11.09	12.77	14.69	15.83	16.85	17.93	19.49
13	14	8.69	10.12	11.64	13.38	15.36	16.52	17.55	18.65	20.21
13.5	3	9.13	10.62	12.20	14.00	16.01	17.19	18.24	19.35	20.92
13.71	0	9.32	10.84	12.44	14.25	16.29	17.48	18.53	19.64	21.22

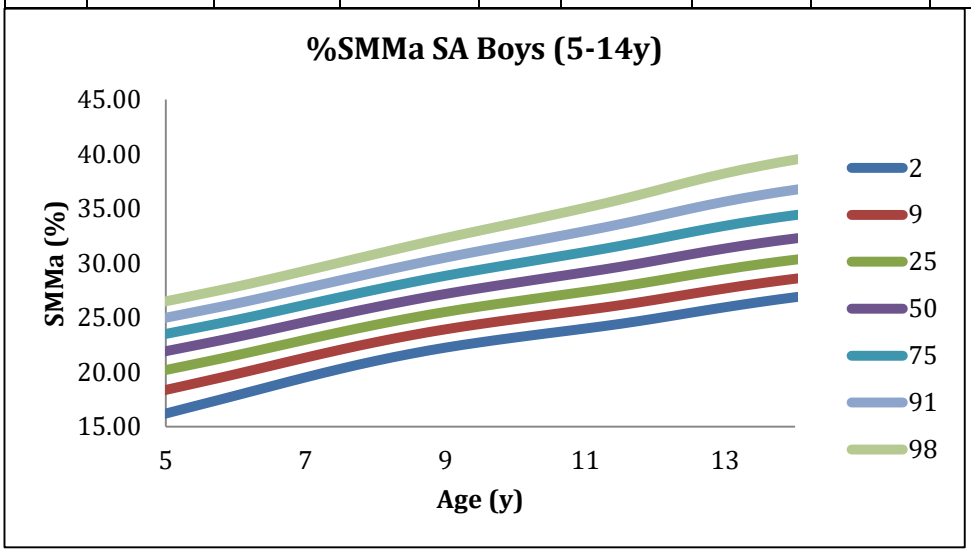



SMMa (kg) centiles - SA & WE2 Boys compared
30
25
20
15
10
5
0
SMMa (kg)
5
6
7
8
9
10
11
12
13
14
Age (y)
SA 2nd centile
SA 50th centile
SA 98th centile
WE 2nd centile
WE 50th centile
WE 98th centile


SMMa (kg) centiles SA & WE2 Girls compared
24
22
20
18
16
14
12
10
8
6
4
2
SMMa (kg)
5
7
9
11
13
Age (y)
SA 2nd centile
SA 50th centile
SA 98th centile
WE 2nd centile
WE 50th centile
WE 98th centile

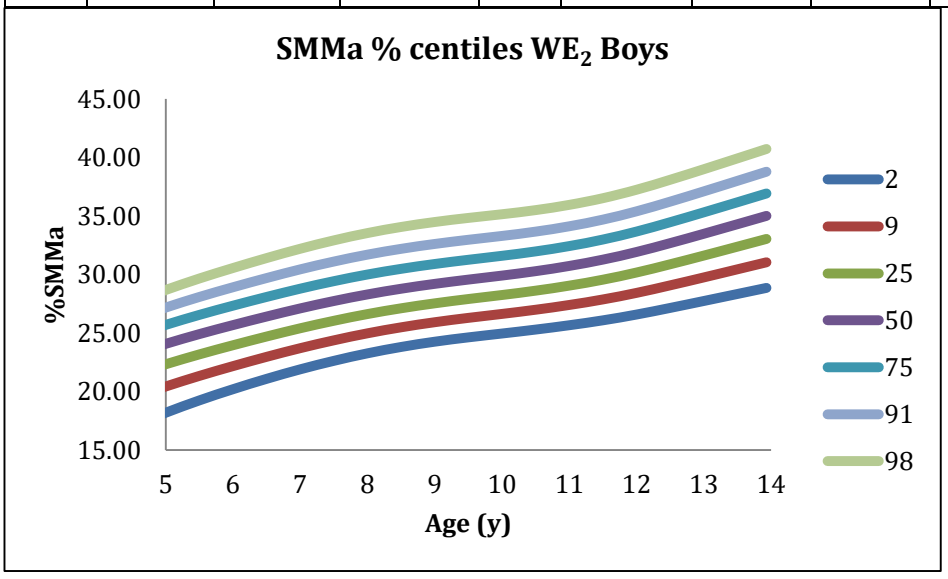
SMMa % Centiles SA Boys

	-2.053749	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
Age	2	9	25	50	75	85	91	95	98
4.29	14.99	17.35	19.26	21.01	22.61	23.41	24.07	24.71	25.53
4.5	15.35	17.64	19.53	21.27	22.87	23.68	24.34	24.98	25.81
5	16.18	18.35	20.19	21.90	23.49	24.31	24.97	25.62	26.48
5.5	17.00	19.07	20.85	22.53	24.12	24.94	25.61	26.27	27.13
6	17.83	19.80	21.53	23.18	24.77	25.59	26.26	26.93	27.80
6.5	18.67	20.56	22.24	23.88	25.46	26.28	26.97	27.64	28.53
7	19.50	21.32	22.96	24.59	26.17	27.00	27.69	28.38	29.29
7.5	20.28	22.05	23.67	25.28	26.87	27.71	28.41	29.11	30.05
8	21.01	22.73	24.33	25.95	27.55	28.41	29.13	29.85	30.81
8.5	21.66	23.36	24.95	26.58	28.21	29.09	29.83	30.57	31.56
9	22.23	23.92	25.52	27.16	28.82	29.73	30.49	31.25	32.29
9.5	22.72	24.40	26.02	27.69	29.39	30.32	31.11	31.90	32.98
10	23.16	24.85	26.48	28.18	29.93	30.89	31.71	32.54	33.66
10.5	23.57	25.27	26.93	28.67	30.47	31.46	32.31	33.17	34.35
11	23.98	25.69	27.37	29.15	31.01	32.04	32.92	33.82	35.06
11.5	24.41	26.13	27.84	29.65	31.57	32.64	33.56	34.51	35.81
12	24.90	26.63	28.35	30.21	32.18	33.29	34.26	35.25	36.62
12.5	25.42	27.15	28.90	30.79	32.82	33.97	34.97	36.01	37.46
13	25.93	27.66	29.42	31.33	33.41	34.60	35.63	36.71	38.22
13.5	26.41	28.14	29.89	31.82	33.94	35.15	36.21	37.33	38.89
14	26.85	28.56	30.31	32.25	34.39	35.62	36.72	37.86	39.48
14.5	27.23	28.93	30.67	32.61	34.77	36.02	37.14	38.31	39.98
15	27.57	29.25	30.98	32.92	35.09	36.36	37.50	38.69	40.41
15.5	27.89	29.54	31.26	33.20	35.38	36.67	37.82	39.05	40.81
15.95	28.18	29.81	31.51	33.45	35.64	36.94	38.11	39.36	41.17



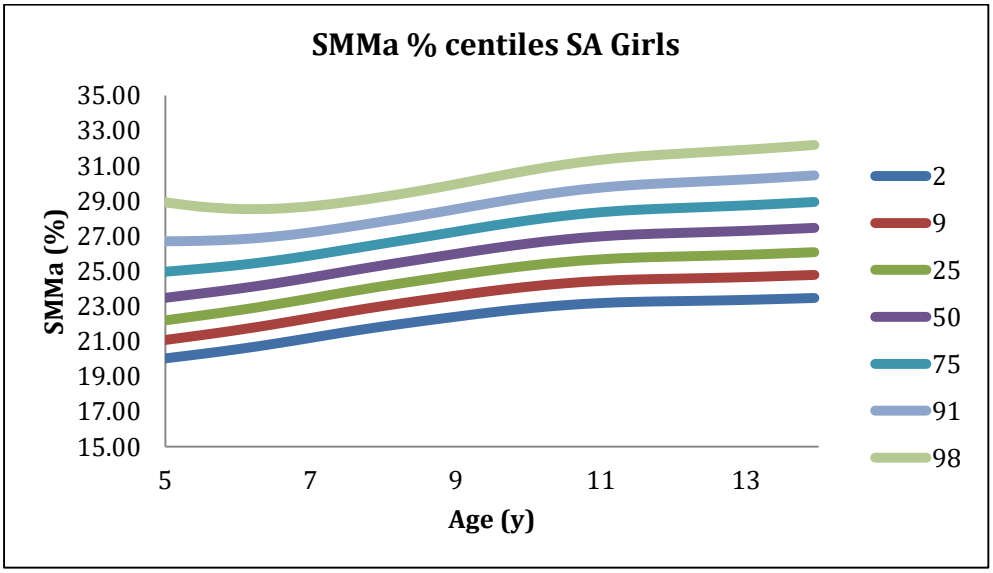
SMMa % centiles WE_2 Boys

Age	-2.053749	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
	2	9	25	50	75	85	91	95	98
4.47	16.99	19.44	21.39	23.14	24.72	25.52	26.16	26.78	27.58
4.5	17.06	19.49	21.44	23.19	24.78	25.57	26.22	26.84	27.65
5	18.19	20.45	22.33	24.08	25.69	26.52	27.19	27.84	28.69
5.5	19.25	21.35	23.17	24.91	26.55	27.40	28.09	28.78	29.67
6	20.20	22.19	23.96	25.68	27.34	28.20	28.92	29.63	30.57
6.5	21.09	22.98	24.70	26.41	28.08	28.97	29.71	30.44	31.42
7	21.90	23.72	25.40	27.10	28.79	29.69	30.45	31.20	32.22
7.5	22.63	24.39	26.05	27.73	29.43	30.35	31.12	31.89	32.93
8	23.27	24.99	26.62	28.30	30.00	30.92	31.71	32.49	33.56
8.5	23.81	25.50	27.11	28.78	30.49	31.41	32.20	33.00	34.08
9	24.26	25.93	27.54	29.20	30.90	31.83	32.62	33.42	34.50
9.5	24.63	26.30	27.90	29.56	31.26	32.19	32.98	33.77	34.85
10	24.96	26.64	28.25	29.91	31.60	32.52	33.30	34.09	35.16
10.5	25.30	27.00	28.62	30.28	31.98	32.90	33.67	34.46	35.52
11	25.67	27.41	29.05	30.73	32.43	33.35	34.13	34.91	35.97
11.5	26.09	27.88	29.56	31.27	32.99	33.92	34.70	35.48	36.54
12	26.58	28.44	30.17	31.92	33.68	34.62	35.41	36.20	37.26
12.5	27.15	29.08	30.87	32.68	34.47	35.43	36.24	37.04	38.11
13	27.74	29.76	31.62	33.48	35.32	36.30	37.11	37.93	39.02
13.5	28.34	30.45	32.38	34.30	36.18	37.18	38.01	38.84	39.94
13.93	28.86	31.05	33.04	35.00	36.93	37.94	38.79	39.63	40.74



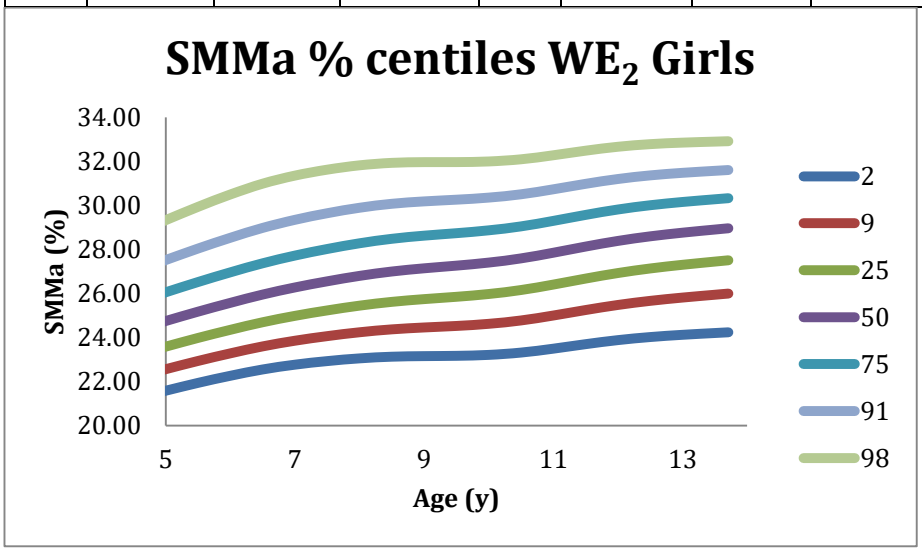
SMMa % centiles SA Girls

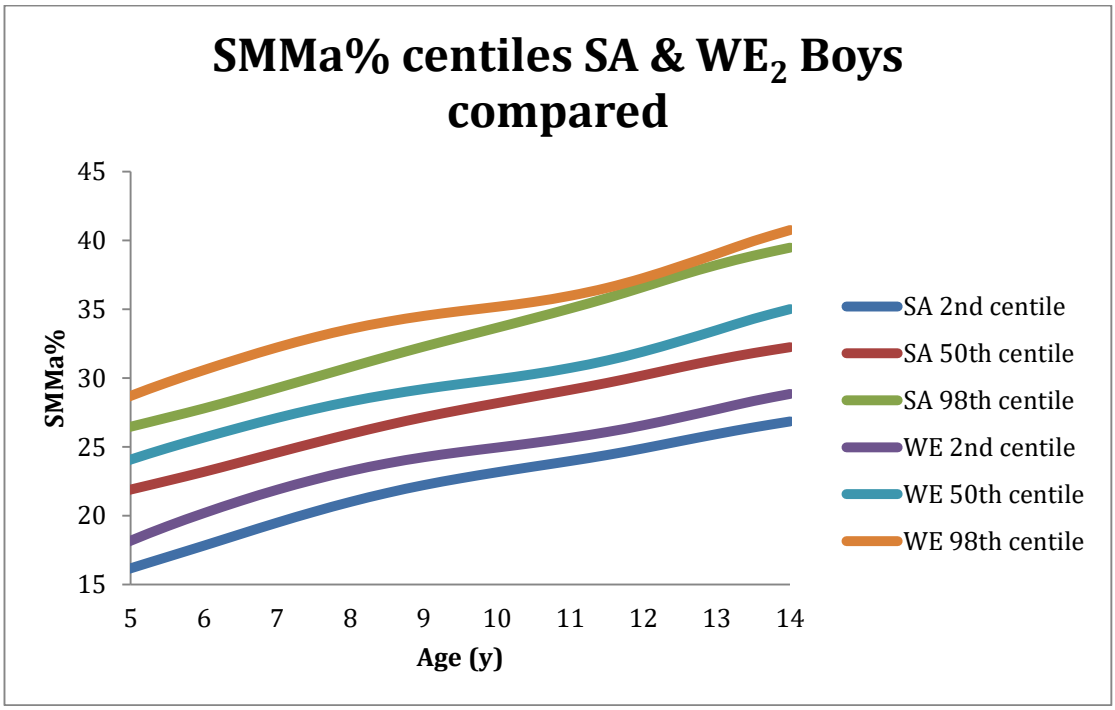
Age	-2.053749	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
	2	9	25	50	75	85	91	95	98
4.46	19.76	20.78	21.89	23.20	24.78	25.76	26.68	27.70	29.28
4.5	19.78	20.81	21.91	23.22	24.79	25.77	26.68	27.69	29.25
5	20.04	21.09	22.20	23.49	24.98	25.88	26.70	27.59	28.92
5.5	20.29	21.37	22.48	23.74	25.15	25.98	26.73	27.52	28.67
6	20.57	21.66	22.77	24.00	25.35	26.12	26.81	27.52	28.54
6.5	20.87	21.98	23.10	24.30	25.60	26.33	26.97	27.63	28.55
7	21.20	22.33	23.45	24.64	25.90	26.60	27.21	27.83	28.69
7.5	21.54	22.69	23.81	25.00	26.23	26.92	27.51	28.11	28.93
8	21.85	23.02	24.15	25.34	26.57	27.25	27.83	28.42	29.23
8.5	22.14	23.32	24.47	25.67	26.91	27.59	28.17	28.76	29.57
9	22.41	23.61	24.77	25.99	27.25	27.94	28.53	29.13	29.95
9.5	22.66	23.88	25.06	26.30	27.58	28.29	28.90	29.51	30.35
10	22.88	24.12	25.32	26.58	27.89	28.62	29.24	29.87	30.73
10.5	23.06	24.31	25.53	26.81	28.15	28.90	29.53	30.18	31.07
11	23.18	24.45	25.68	26.99	28.35	29.11	29.76	30.42	31.34
11.5	23.25	24.53	25.78	27.10	28.49	29.26	29.92	30.60	31.53
12	23.29	24.57	25.84	27.17	28.58	29.36	30.03	30.72	31.67
12.5	23.32	24.61	25.89	27.24	28.66	29.45	30.13	30.83	31.79
13	23.36	24.66	25.94	27.31	28.74	29.54	30.23	30.94	31.91
13.5	23.42	24.73	26.02	27.39	28.84	29.65	30.35	31.06	32.05
13.94	23.47	24.79	26.09	27.47	28.94	29.75	30.46	31.18	32.18

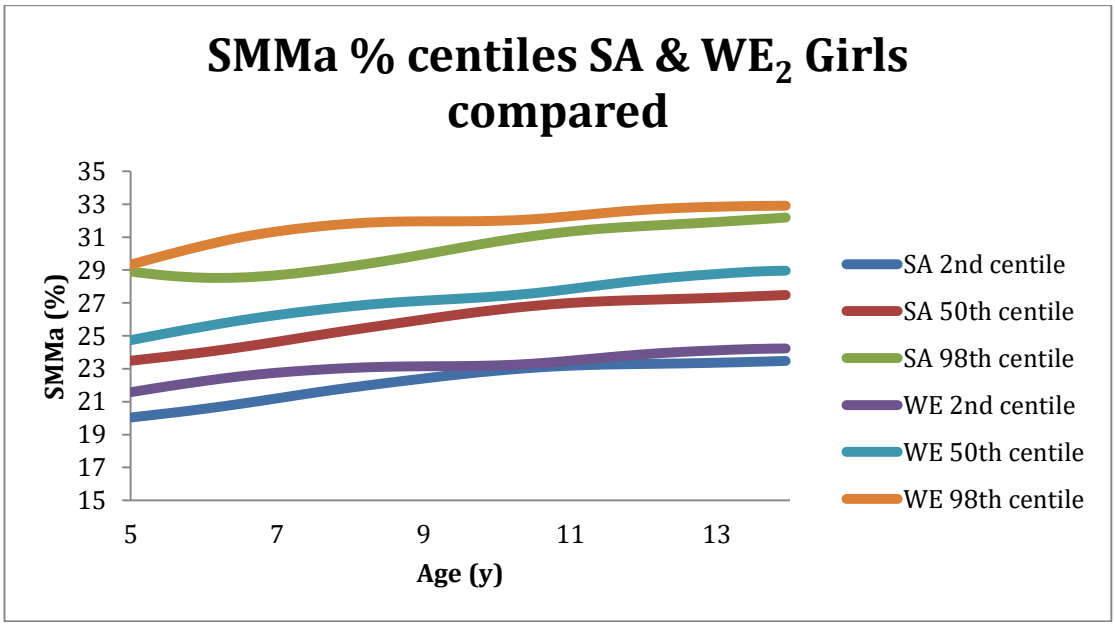


SMMa % centiles WE_2 Girls

Age	-2.053749	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
	2	9	25	50	75	85	91	95	98
4.51	21.22	22.19	23.19	24.32	25.58	26.33	27.00	27.70	28.73
5	21.58	22.56	23.58	24.74	26.05	26.82	27.51	28.25	29.33
5.5	21.93	22.93	23.98	25.17	26.51	27.31	28.04	28.81	29.93
6	22.26	23.28	24.35	25.57	26.95	27.78	28.53	29.32	30.49
6.5	22.55	23.60	24.69	25.94	27.36	28.21	28.97	29.79	30.98
7	22.76	23.85	24.98	26.27	27.72	28.57	29.34	30.16	31.35
7.5	22.93	24.06	25.23	26.55	28.01	28.87	29.64	30.45	31.62
8	23.05	24.24	25.45	26.79	28.27	29.13	29.89	30.69	31.82
8.5	23.13	24.37	25.62	26.99	28.48	29.33	30.07	30.84	31.93
9	23.15	24.45	25.75	27.14	28.62	29.46	30.18	30.93	31.96
9.5	23.17	24.52	25.85	27.26	28.73	29.55	30.26	30.98	31.96
10	23.21	24.62	25.98	27.40	28.86	29.67	30.35	31.05	31.99
10.5	23.32	24.77	26.16	27.59	29.04	29.83	30.50	31.18	32.09
11	23.49	25.00	26.41	27.85	29.29	30.07	30.73	31.39	32.27
11.5	23.70	25.24	26.68	28.13	29.56	30.33	30.98	31.62	32.48
12	23.88	25.47	26.93	28.38	29.81	30.57	31.20	31.82	32.66
12.5	24.02	25.66	27.14	28.59	30.00	30.74	31.36	31.97	32.78
13	24.12	25.81	27.31	28.76	30.15	30.88	31.48	32.07	32.85
13.5	24.21	25.94	27.45	28.90	30.27	30.98	31.57	32.14	32.89
13.71	24.24	25.99	27.51	28.96	30.32	31.03	31.60	32.17	32.91

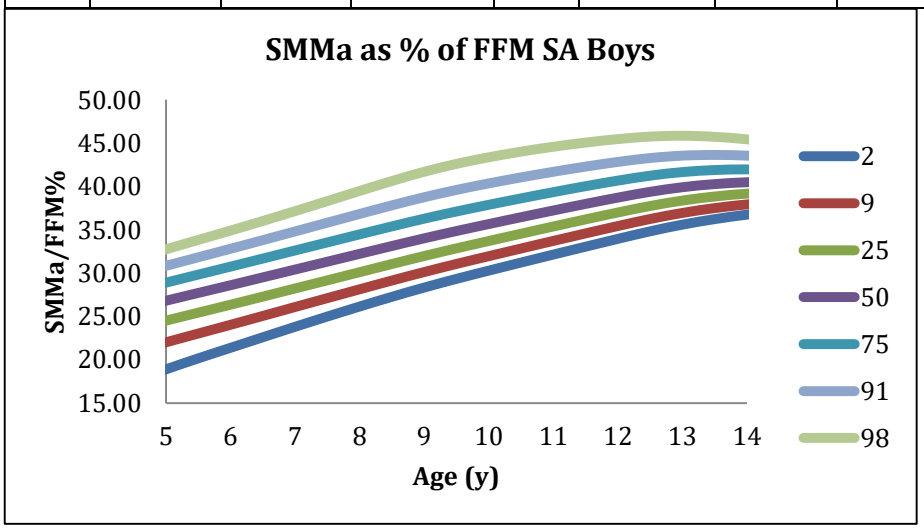



SMMa% centiles SA & WE2 Boys compared
45
40
35
30
25
20
15
SMMa%
5
6
7
8
9
10
11
12
13
14
Age (y)
SA 2nd centile
SA 50th centile
SA 98th centile
WE 2nd centile
WE 50th centile
WE 98th centile


SMMa % centiles SA & WE2 Girls compared
35
33
31
29
27
25
23
21
19
17
15
SMMa (%)
5
7
9
11
13
Age (y)
SA 2nd centile
SA 50th centile
SA 98th centile
WE 2nd centile
WE 50th centile
WE 98th centile

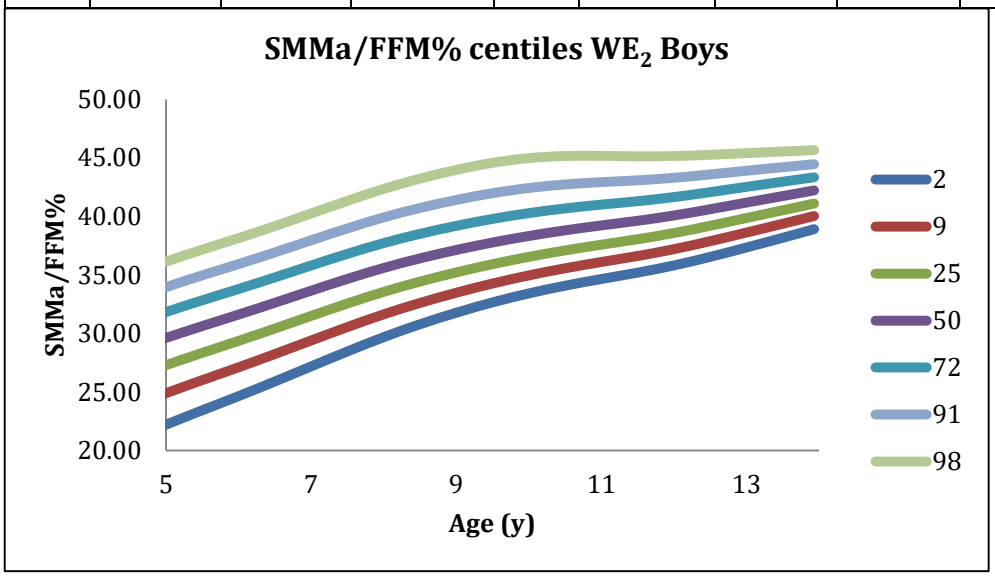
SMMa % of FFM SA Boys

Centiles									
	-2.053749	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
Age	2	9	25	50	75	85	91	95	98
4.29	16.92	20.54	23.22	25.54	27.58	28.59	29.39	30.17	31.17
4.5	17.52	20.97	23.61	25.91	27.97	28.99	29.82	30.61	31.63
5	18.88	22.00	24.52	26.81	28.91	29.96	30.82	31.65	32.73
5.5	20.15	23.02	25.44	27.71	29.83	30.92	31.81	32.68	33.81
6	21.37	24.03	26.36	28.61	30.75	31.87	32.79	33.70	34.89
6.5	22.59	25.05	27.29	29.51	31.68	32.83	33.79	34.74	36.01
7	23.79	26.08	28.23	30.42	32.62	33.80	34.80	35.80	37.15
7.5	24.98	27.11	29.17	31.33	33.55	34.77	35.81	36.87	38.31
8	26.14	28.13	30.11	32.23	34.48	35.73	36.82	37.93	39.48
8.5	27.26	29.14	31.05	33.14	35.40	36.69	37.82	38.98	40.63
9	28.33	30.13	31.97	34.03	36.29	37.60	38.76	39.97	41.69
9.5	29.33	31.06	32.86	34.87	37.11	38.43	39.60	40.84	42.61
10	30.28	31.96	33.71	35.68	37.89	39.19	40.35	41.59	43.37
10.5	31.21	32.85	34.54	36.47	38.63	39.90	41.05	42.26	44.02
11	32.13	33.72	35.38	37.24	39.35	40.58	41.70	42.88	44.59
11.5	33.05	34.59	36.20	38.00	40.03	41.23	42.30	43.43	45.07
12	33.95	35.44	36.99	38.73	40.67	41.82	42.84	43.91	45.47
12.5	34.82	36.26	37.74	39.40	41.24	42.32	43.29	44.30	45.76
13	35.59	36.96	38.36	39.93	41.66	42.67	43.56	44.51	45.86
13.5	36.23	37.52	38.83	40.29	41.89	42.82	43.64	44.51	45.74
14	36.74	37.95	39.16	40.51	41.97	42.82	43.56	44.34	45.44
14.5	37.14	38.25	39.36	40.58	41.91	42.66	43.33	44.02	45.00
15	37.45	38.45	39.46	40.55	41.72	42.39	42.98	43.58	44.44
15.5	37.70	38.59	39.48	40.45	41.47	42.06	42.56	43.08	43.82
15.95	37.91	38.70	39.49	40.34	41.23	41.74	42.17	42.62	43.25



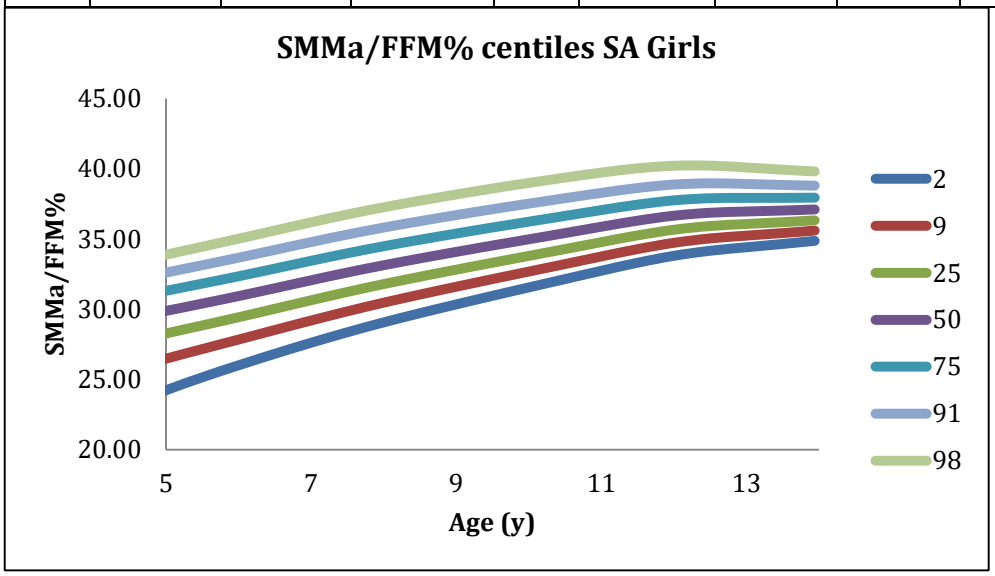
SMMa/FFM% centiles WE_2 Boys

	Centiles								
Age	-2.053749	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
	2	9	25	50	72	85	91	95	98
4.47	20.85	23.68	26.14	28.48	30.70	31.85	32.80	33.72	34.94
4.5	20.93	23.75	26.20	28.54	30.77	31.91	32.86	33.79	35.01
5	22.19	24.90	27.29	29.60	31.82	32.98	33.94	34.88	36.12
5.5	23.43	26.01	28.34	30.62	32.83	33.99	34.95	35.90	37.16
6	24.63	27.11	29.36	31.60	33.79	34.95	35.91	36.87	38.14
6.5	25.88	28.24	30.42	32.61	34.78	35.94	36.91	37.88	39.17
7	27.17	29.40	31.50	33.64	35.80	36.96	37.94	38.93	40.25
7.5	28.45	30.54	32.56	34.65	36.79	37.96	38.95	39.96	41.33
8	29.67	31.63	33.55	35.58	37.71	38.89	39.90	40.93	42.35
8.5	30.77	32.60	34.43	36.40	38.49	39.68	40.70	41.76	43.23
9	31.76	33.48	35.21	37.11	39.18	40.36	41.39	42.46	43.98
9.5	32.64	34.26	35.92	37.75	39.77	40.94	41.97	43.05	44.58
10	33.41	34.96	36.54	38.31	40.26	41.40	42.41	43.47	44.98
10.5	34.07	35.56	37.09	38.79	40.67	41.75	42.71	43.72	45.16
11	34.64	36.10	37.58	39.21	40.99	42.01	42.91	43.85	45.18
11.5	35.18	36.62	38.05	39.61	41.28	42.24	43.07	43.93	45.14
12	35.80	37.20	38.58	40.06	41.63	42.52	43.28	44.07	45.16
12.5	36.53	37.88	39.20	40.60	42.06	42.87	43.57	44.29	45.27
13	37.33	38.62	39.86	41.16	42.51	43.25	43.88	44.53	45.41
13.5	38.16	39.37	40.52	41.72	42.95	43.62	44.19	44.76	45.55
13.93	38.90	40.02	41.10	42.20	43.32	43.93	44.45	44.96	45.67



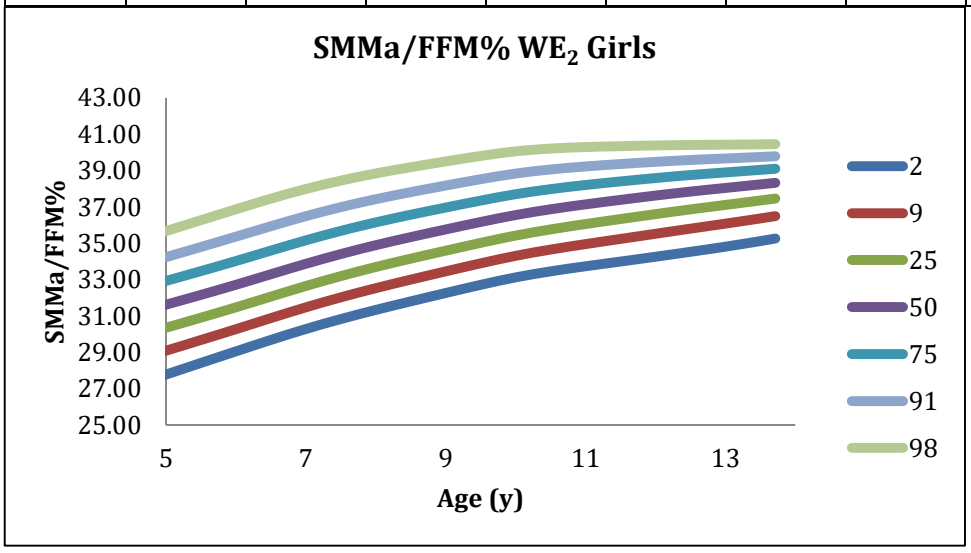
SMMa/FFM% centiles SA Girls

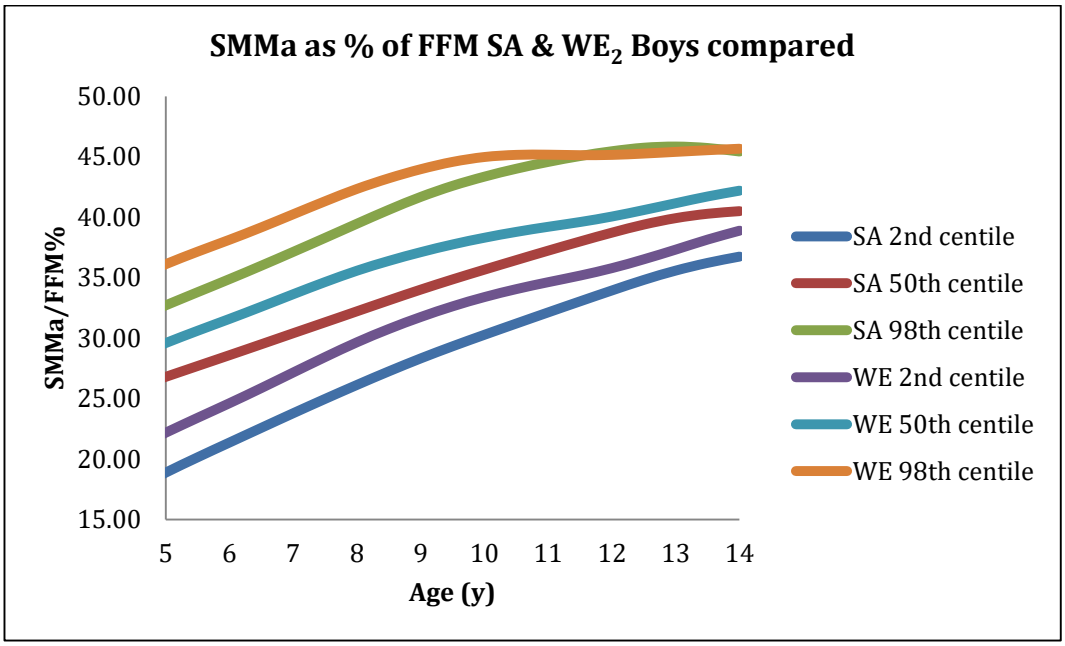
	Centiles								
Age	-2.053749	-1.340755	-0.6744897	0	0.6744897	1.036433	1.340755	1.644854	2.053749
	2	9	25	50	75	85	91	95	98
4.46	23.10	25.70	27.64	29.29	30.74	31.45	32.01	32.55	33.25
4.5	23.19	25.76	27.68	29.34	30.78	31.49	32.06	32.60	33.29
5	24.22	26.47	28.27	29.87	31.30	32.02	32.60	33.16	33.88
5.5	25.14	27.16	28.84	30.39	31.82	32.54	33.13	33.70	34.45
6	26.00	27.83	29.42	30.92	32.34	33.07	33.67	34.25	35.02
6.5	26.82	28.52	30.02	31.49	32.89	33.62	34.22	34.82	35.60
7	27.60	29.19	30.63	32.05	33.44	34.17	34.78	35.38	36.18
7.5	28.35	29.85	31.23	32.61	33.98	34.71	35.32	35.92	36.74
8	29.05	30.46	31.78	33.13	34.48	35.20	35.81	36.43	37.25
8.5	29.71	31.04	32.31	33.61	34.94	35.66	36.27	36.89	37.72
9	30.33	31.60	32.81	34.08	35.38	36.09	36.70	37.32	38.16
9.5	30.94	32.14	33.31	34.53	35.81	36.52	37.12	37.74	38.59
10	31.54	32.68	33.79	34.98	36.22	36.92	37.52	38.13	38.98
10.5	32.14	33.21	34.28	35.42	36.63	37.32	37.91	38.52	39.36
11	32.73	33.75	34.77	35.87	37.05	37.71	38.29	38.88	39.72
11.5	33.30	34.28	35.25	36.31	37.44	38.08	38.64	39.21	40.02
12	33.79	34.73	35.66	36.66	37.74	38.35	38.88	39.43	40.20
12.5	34.15	35.04	35.92	36.87	37.89	38.47	38.97	39.49	40.21
13	34.40	35.24	36.07	36.96	37.91	38.45	38.92	39.40	40.08
13.5	34.64	35.42	36.20	37.03	37.92	38.42	38.85	39.30	39.92
13.94	34.86	35.59	36.32	37.11	37.93	38.40	38.80	39.22	39.80

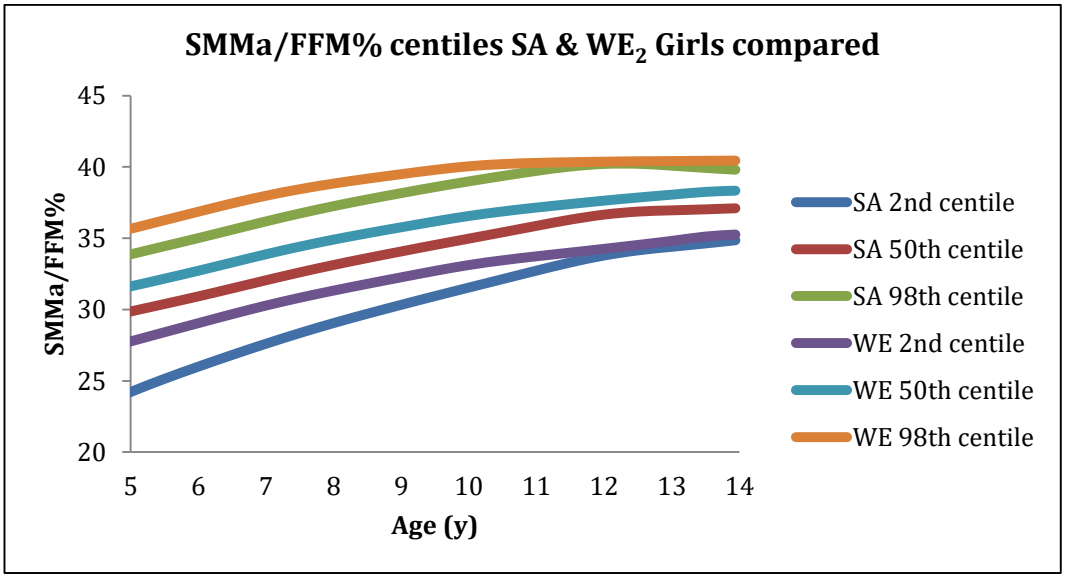


SMMa/FFM% centiles WE_2 Girls

	Centiles								
Age	-2.05	-1.34	-0.67	0.00	0.67	1.04	1.34	1.64	2.05
	2	9	25	50	75	85	91	95	98
4.51	27.14	28.52	29.81	31.11	32.41	33.11	33.70	34.28	35.07
5	27.77	29.09	30.34	31.63	32.93	33.64	34.24	34.84	35.66
5.5	28.41	29.68	30.90	32.16	33.47	34.19	34.80	35.42	36.26
6	29.05	30.27	31.46	32.72	34.02	34.75	35.36	35.99	36.86
6.5	29.68	30.88	32.06	33.30	34.60	35.32	35.94	36.57	37.45
7	30.28	31.47	32.64	33.87	35.16	35.88	36.49	37.12	37.98
7.5	30.83	32.02	33.19	34.41	35.68	36.38	36.99	37.60	38.44
8	31.33	32.53	33.69	34.90	36.15	36.84	37.42	38.02	38.84
8.5	31.80	33.00	34.15	35.34	36.57	37.24	37.81	38.39	39.18
9	32.26	33.45	34.59	35.76	36.96	37.62	38.17	38.74	39.50
9.5	32.71	33.89	35.02	36.17	37.35	37.98	38.52	39.07	39.80
10	33.12	34.30	35.42	36.56	37.70	38.31	38.83	39.35	40.05
10.5	33.45	34.65	35.76	36.87	37.98	38.56	39.06	39.55	40.21
11	33.73	34.95	36.05	37.14	38.20	38.76	39.22	39.68	40.29
11.5	33.99	35.23	36.33	37.39	38.40	38.92	39.36	39.78	40.34
12	34.25	35.52	36.60	37.63	38.58	39.08	39.48	39.87	40.38
12.5	34.53	35.80	36.86	37.85	38.75	39.21	39.58	39.94	40.41
13	34.81	36.07	37.11	38.05	38.89	39.32	39.66	39.99	40.42
13.5	35.12	36.36	37.35	38.24	39.03	39.43	39.74	40.05	40.44
13.71	35.25	36.48	37.46	38.32	39.09	39.48	39.78	40.08	40.45




SMMa as % of FFM SA & WE2 Boys compared
50.00
45.00
40.00
35.00
30.00
25.00
20.00
15.00
SMMa/FFM%
5
6
7
8
9
10
11
12
13
14
Age (y)
SA 2nd centile
SA 50th centile
SA 98th centile
WE 2nd centile
WE 50th centile
WE 98th centile


SMMa/FFM% centiles SA & WE2 Girls compared
45
40
35
30
25
20
SMMa/FFM%
5
6
7
8
9
10
11
12
13
14
Age (y)
SA 2nd centile
SA 50th centile
SA 98th centile
WE 2nd centile
WE 50th centile
WE 98th centile