Title The Validity of Two Compartment Model Methods of Body Composition as Compared to Magnetic Resonance Imaging in Asian Indian Versus Caucasian Males

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The validity of 2 compartment model methods of body composition as compared to magnetic resonance imaging in Asian Indian versus Caucasian males.

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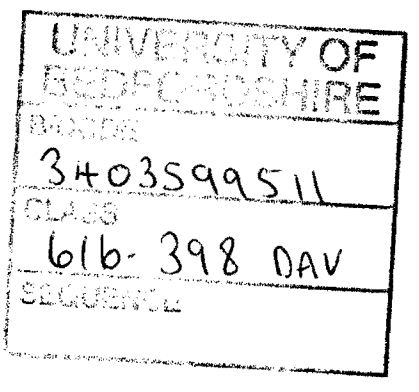

DECLERATION

I declare that this thesis is my own unaided work. It is being submitted for the degree of MSc by research at the University of Bedfordshire.

It has not been submitted before for any degree or examination in any other University.

Name of candidate: **Ben Davies** Signature:

Date: 26/01/2010

ABSTRACT

Background: The two-compartment (2C) model is a relatively accessible, inexpensive and time efficient method for body composition measurement. There is very little validated research on the 2C model in Asian Indians: a high risk population in terms of obesity and related disorders. This highlights the need for valid estimates of body composition from the 2C model. **Purpose:** The goal was to compare 2C model (predictor) estimates of body composition to those from magnetic resonance imaging (MRI) (criterion), an established gold standard measure of total adiposity in order to determine the validity of the 2C model in the Asian Indian population. From this data it is hoped that a correction equation may be determined for more accurate prediction of Asian Indian body composition using 2C model methods. **Methods:** 21 males (10 Asian Indian and 11 Caucasian, aged 18-55 yrs) had estimates of percent body fat from 2C methods (sum of four skinfolds and anthropometry, bioelectrical impedance analysis [Bodystat 1500 and Tanita segmental impedance analyser], air displacement plethysmography [BodPod] and hydrostatic weighing) compared to MRI measured body composition values. Agreement was assessed using multiple linear regression analysis and Bland-Altman plots. Differences were assessed using repeated measures analysis of variance. **Results:** Regression analysis showed air displacement plethysmography predicts MRI body composition in Caucasian males (adjusted r^2 = 0.74; SEE = 3.27). In Asian Indians, tricep skinfold thickness and hydrostatic weighing predicted MRI body composition with a low prediction error (adjusted r^2 = 0.90; SEE = 1.75). Despite strong correlations and no significant difference between mean differences of the 2C methods, used in the prediction model, and MRI, Bland-Altman plots revealed no acceptable limits of agreement between the methods. Asian Indian body composition was underestimated by all two compartment devices compared to MRI. **Conclusion:** There appears to be potential for the use of tricep skinfold thickness and hydrostatic weighing to predict an established reference measure (MRI) in the high risk Asian Indian population. The 2C model underestimated Asian Indian body composition, this suggests that un-validated, the 2C model may misidentify obesity and in turn health risk. However the small sample tested, has implications for the interpretation of the findings. Further investigation is required with a greater sample size to validate the 2C model against an established reference measure such as MRI as there is currently little published validation data in this ethnic group.

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LIST OF ABBREVIATIONS

ADP - Air displacement plethysmography

BIA – Bioelectrical impedance analysis

CT – Computerised tomography

CV – Coefficient of variation

Db – Body density

DXA – Dual energy X-ray absorptiometry

FFB – Fat free body

FFM – Fat free mass

FM – Fat mass

FVC – Forced vital capacity

HW – Hydrostatic weighing

LTM – Lean tissue mass

NAA – Neutron activation analysis

NEFA – Non esterised fatty acids

r – Correlation coefficient

r^2 - Coefficient of determination

RLV – Residual lung volume

SEE – Standard error of the estimate

STM – Soft tissue mass

TBBW – Total body bone mineral

TBW – Total body mineral

TVG – Thoracic gas volume

2C – Two-compartment

1. INTRODUCTION

Obesity (an excessive amount of total body fat relative to body weight) is a major concern affecting individuals across the world with problems first identified in the 1970's in a number of government reports including the National Study of Health and Growth in 1974-1994 and in the 1995 Health Survey for England. These reports demonstrated that the prevalence of obesity in the British population had sharply increased from 6% to 17% in males, and 8% to 21% in females. More worryingly, these reports indicated, that among British children, the prevalence had tripled from 0.6% to 1.7% in boys and from 1.3% to 2.6% in girls.

The spread of obesity is apparent from the World Health Organisation (WHO) 2002 Obesity Report, which monitors trends and prevalence of obesity, states that the majority of European countries have shown a 10-40% increase in obesity rates between 1987 and 2002, and furthermore England demonstrated the most dramatic increase of over 100% (WHO, 2002). The WHO (2002) state that obesity has become a global epidemic, the dramatic increase no longer only exists in affluent communities, but is now evident in areas of low socio-economic status, in developing countries, such as China, India and Thailand (Caballero 2007). The WHO (2005) projected there to be approximately 1.6 billion overweight adults (aged 15+) and at least, 20 million overweight children (< 5 years of age), globally, of the adults 400 million of those individuals may be classified as obese. The WHO forecasts, that by 2015 2.3 billion will be overweight

with 700 million obese, showing a predicted 50% increase in the prevalence of obesity in the intervening 10 years.

This dramatic increase in worldwide obesity is multifactorial. Altered eating habits and expanded food options have led to increased production of readily available, high fat, energy dense foods (Hill et al, 2000). Added to this, more people are sedentary as a result of advances in technology, such as mechanisation and automation (Hill et al, 2000). A contributor to fat gain is the combination of high energy intake and low energy output, which produces a positive energy balance. This excess energy is stored in the body promoting weight gain (WHO, 2004). Studies have shown that genetic factors play a major role in the regulation of body weight (Hill et al, 2000). However, the recent rapid increase in obesity suggests environmental factors and lifestyle choices must be largely to blame.

The disorders secondary to obesity, such as type II diabetes and cardiovascular disease, are strongly linked to high morbidity and mortality rates (Bhat et al, 2005; Raji et al, 2001). Type 2 diabetes has rapidly become a pandemic, projected by WHO, to increase by 50% between 2005 and 2015 (WHO, 06). Cardiovascular disease (most commonly heart disease and stroke) is already the leading cause of mortality in the world (WHO, 2006). Others include: hypertension (high blood pressure; Rönnback et al, 2007), dyslipidemia (e.g. high total cholesterol; Rönnback et al, 2007), and osteoarthritis (breakdown of bone and cartilage within a

joint, due to excessive load; Shedd et al, 2007). It is without doubt that obesity and life threatening disease are strongly linked.

Research has shown that calorie restricted diet and exercise can reduce obesity and its associated health risks (Ross et al, 2000; Blair et al 1989). In the last decade the British government has invested £372 million on weight management implementation. According to the Department of Health (2008), obesity currently costs the British National Health Service (NHS) an estimated £4.2 billion per year which is expected to more than double by the year 2050. These figures clearly demonstrate that obesity is an economic as well as public health problem.

Early studies used body mass index (BMI), utilising simple measures of height and weight to classify obese, overweight and underweight; it was traditionally the most common method in research. BMI, however, doesn't account for the greater weight of muscle mass than fat and thus can miss-classify individuals as too heavy for their height, categorising them as overweight or obese when they may be very lean. Although research has shown that BMI is a significant predictor of type 2 diabetes and cardiovascular disease, it is not as accurate on a more individual basis (Janssen et al, 2002). By measuring body composition, it can be clearly determined whether an individual is overweight or obese. It has recently become apparent that not only overall body composition but fat deposition is important for identification of obesity related risk. Distribution of fat, abdominal and visceral fat in particular, have recently become factors for

concern due to their association with obesity related disorders (Bacha et al, 2003). Abdominal obesity is a risk factor constituting to the metabolic syndrome, further to this, other factors include diabetes, high cholesterol and hypertension. The metabolic syndrome has been defined in adults as a cluster of the most dangerous risk factors for type II diabetes and cardiovascular disease (Alberti et al, 2005). Distribution of fat is known to vary between different ethnic groups. Certain ethnicities store more fat centrally than others; Blacks less than Caucasian less than Southern Asians (Schutte et al al, 1984; Chandalia et al, 1999). The issue of ethnicity and body fat is highlighted further by the fact that recently, new BMI cut points were proposed for Asian Indians (WHO, 2004), suggesting that for a number of years, obesity related risk has been underestimated without the correct guidance or treatment. This could be a factor linked to the epidemic proportions of type II diabetes seen in Asian Indians today (Ramachandran et al, 2001).

The two-compartment (2C) model is the most commonly used approach of measuring body composition. It assumes the body is made of two compartments, a fat compartment and fat free compartment, and assumes constant densities of fat and fat free tissue to estimate body fat. It is used widely in domestic and clinical settings as well as in the fitness industry to monitor body fat levels. The equations used to estimate body composition include standardised algorithms based on the constant densities of fat and fat free mass. These algorithms were derived from limited reference cadaver data (Siri, 1956; Brozek et al, 1963) of a small number of White

Caucasians. These algorithms may not apply specifically to non-white ethnic groups. Research (Schutte et al, 1984) has revealed that different ethnic groups demonstrate different fat free mass densities, thus producing inaccurate body fat values when measured using the 2C model. This has led to the re-investigation of the 2C model on different ethnic groups, and the development of new equations for black individuals (Schutte et al, 1984) enhancing the accuracy of their body composition measures. There is very limited evidence of the validity of the 2C model on Asians and in particular southern Asian Indians. The importance of investigating the 2C model's validity in this ethnic group is highlighted by evidence that Asian Indians are a high risk population in terms of obesity related disorders and, in turn, morbidity and mortality, as they are known to store a greater amount of centralised fat (Banerji et al, 1999; Raji et al, 2001). Understanding the validity of the 2C model in this ethnic group will improve our ability to identify obesity and determine potential risks to health as it is the most prevalent approach to measuring body composition. Therefore, in order to assess the accuracy of the 2C model in Asian Indians the 2C model will be compared to an established criterion, magnetic resonance imaging (MRI) with the aim of developing a correction factor to improve the accuracy of the 2C model if required, for Southern Asian Indians.

Aim

The current research aims to quantify the validity of established, simple 2C methods of estimating body composition against a criterion method in

Asian Indian males. Evidence of ethnic variation in fat free mass density warrants the use of a criterion measure that does not assume the density of the fat free body when measuring body composition in Asian Indians. Therefore 2C methods of body composition will be compared to MRI. The main objective of this investigation is to elucidate the accuracy of simple methods used to estimate body composition in Asians Indians, thus allowing for more accurate identification of potential health risk, using the following research question: Can a correction factor be developed for more accurate estimation of Asian male adiposity from common methods of the two-compartment model?

Hypotheses

Based on previous literature the following null ($\mathbf{H^0}$) and alternate ($\mathbf{H^1}$) hypotheses have postulated:

1. **H^0:** There will be no significant difference between percent body fat values estimated by the 2C model methods (ADP, BIA, HW and SKF) compared to the criterion MRI.

   H^1: There will be a significant difference between percent body fat values estimated by the 2C model methods (ADP, BIA, HW and SKF) compared to the criterion MRI.

2. **H^0:** There will be no significant linear relationship between percent body fat scores estimated by the 2C model methods (ADP, BIA, HW and SKF) compared to the criterion MRI.

H^1: There will be a significant linear relationship between percent body fat scores estimated by the 2C model methods (ADP, BIA, HW and SKF) compared to the criterion MRI.

3.

H^0: There will not be sufficient agreement to infer validity between percent body fat scores estimated by the 2C model methods (ADP, BIA, HW and SKF) compared to the criterion MRI.

H^1: There will be sufficient agreement to infer validity between percent body fat scores estimated by the 2C model methods (ADP, BIA, HW and SKF) compared to the criterion MRI.

2. REVIEW OF LITERATURE

Obesity is a health risk

The threat to health from obesity is well recognised in literature, and importantly so due to the high morbidity and mortality rates associated with the diseases secondary to obesity (WHO, 2004). Of greatest concern are diabetes, cardiovascular disease and a number of cancers (endometrial, colon and breast). Type II diabetes is currently the most researched obesity related disorder. Due to its recent and extremely rapid growth, it has become a global epidemic, projected to increase 50% by 2015 (WHO, 2006). Evidence that cardiovascular disease (most commonly heart disease and stroke) is the leading cause of mortality in the world (WHO, 2006) highlights the threat of obesity further. Other disorders linked to obesity are hypertension, high cholesterol and osteoarthritis. All are a huge burden to those diagnosed, contributing to a deterioration in health and, potentially, death (WHO, 2005).

Obesity and fat distribution

Understanding obesity levels and the distribution of body fat is an important consideration when assessing health risk. It is understood that excess body fat stored around the abdominal region (central obesity) is associated with disease (Bacha et al, 2003; Banerji et al, 1999; Chandalia et al, 1999; Peris et al, 1989). Although evidence would suggest a clear association with central obesity and health risk, there is conflicting

evidence as to what compartment of fat is most highly associated with disease.

A number of studies have examined the importance of adiposity and fat distribution to cardiovascular risk profile. Peiris et al (1989) measured body composition, fat distribution (measured by computed axial tomography [CT]), insulin response and blood and lipid profile as factors to determine cardiovascular risk of 33 healthy, premenopausal women. Observations showed that intra-abdominal fat (visceral fat in the abdominal cavity; Hayward & Wagner 2004) accounted for a significantly greater degree of variance in cardiovascular risk factors than total body fat. These findings suggest that it is more important to determine the distribution of fat rather than just total body fat alone. They observed that cardiovascular disease may primarily be caused by hyperinsulinemia (increased levels of insulin in the body due to type II diabetes), secondary to intra-abdominal obesity. However the statistical association between hyperinsulinemia, as a result of abdominal obesity, and the inducement of cardiovascular disease cannot confirm causality. This highlights the need for further investigation into the mechanisms by which fat distribution predisposes patients to metabolic problems. Increased visceral fat deposition has been linked to metabolic and cardiovascular risk factors such as insulin resistance, type II diabetes, an adverse lipid profile and cardiovascular disease (Banerji et al, 1999). Banerji et al, (1999) interestingly reported an increased waist to hip ratio was linked to insulin resistance in obese but not lean Asian Indians and that total body fat was positively correlated with insulin resistance in lean Asian Indians. This

suggests that visceral fat may be the prime contributor to cardiovascular risk factors.

The link between central obesity, type II diabetes and cardiovascular disease has also been reported in females by Caprio et al, (1993) who used MRI to examine fat distribution and cardiovascular risk factors in adolescent girls; 14 were obese and 10 were non-obese. Participants had height, weight, waist circumference and hip circumference measured. MRI was used to directly obtain intra abdominal fat deposition. From this, total and visceral abdominal fat and subcutaneous fat were calculated. Central fat storage was twice to three times greater in obese compared to non-obese girls, demonstrated by greater waist to hip ratios in the obese group. MRI showed that, specifically, visceral fat had a significant positive correlation with metabolic risk factors linked to type II diabetes and cardiovascular risk factors. It is important to note the use of imaging techniques such as CT (Peris et al, 1989; Banerji et al, 1999) and MRI scanning (Caprio et al, 1993). These techniques are required for accurate quantification of fat below the surface of the skin and deep within the abdomen to allow for levels of internal fat to be evaluated.

Valsamakis et al, (2004) also used MRI and simple anthropometric measures of obesity to investigate the relationship between visceral fat and the metabolic syndrome using a sample of 46 type II diabetics and 37 non diabetic males, matched for BMI. Their main findings suggest that

those who were diabetic demonstrated an increased generalised abdominal (subcutaneous and visceral) obesity.

It is not entirely clear as to which compartments of fat are the greatest contributors to cardiovascular risk factors. However, it is evident that central obesity is linked with potentially life threatening health risk. Evidence would suggest that perhaps visceral fat has a stronger association with cardiovascular risk factors than general centralised adiposity as a whole (Peris et al, 1989; Bacha et al 2003).

Fat deposition and ethnicity

Recent evidence (Abate et al, 2004; Bacha et al 2003; He at al, 2002) has shown that different ethnic groups store body fat differently. With the understood threat of central obesity, research has investigated the variation of fat deposition in different ethnic groups to identify those who may be at greater risk.

Bacha et al (2003) investigated obesity and regional fat distribution and its association with metabolic risk factors in Black (12 male and 12 female) compared to White (14 male and 12 female), obese adolescents. Fifty participants were examined, comprised of 24 Black (Afro American) and 26 White obese adolescents (mean age 13.35 ± 0.35 years). Insulin sensitivity and secretion were measured as well as lipid profile and blood pressure. Body composition was determined by dual-energy X-ray absorptiometry (DXA) and subcutaneous abdominal and visceral adipose

tissue were examined via CT scan at L4-L5. Both groups had similar age, BMI and body composition. CT scans revealed Whites had 30% more visceral fat than blacks. The authors propose this may be the reason why blacks demonstrated a better lipid profile than that associated with greater atherogenic risk observed in Whites. Despite lower visceral fat in Blacks, insulin sensitivity was not higher. This can be related to evidence that Blacks in general have lower insulin sensitivity than Whites (Arslanian, 2002). Bacha et al (2003) concluded that obese Blacks had significantly less visceral fat than obese Whites and that this was related to increased metabolic and cardiovascular risk factors in Whites. Visceral fat was associated with cardiovascular and metabolic risk in both groups but ethnicity moderated different responses to visceral fat.

Increased visceral fat accumulation in Whites compared to Blacks was also reported by Goran et al (1997) who investigated visceral fat in White (16 males and 20 females) and African American (27 males and 38 females) prepubertal boys and girls (mean age 7.7 ± 1.6 years), determined via CT taken at the level of the umbilicus. The authors observed a wide variation in visceral fatness between the two groups. However, once an index of visceral fat independent of fat mass (FM) was aquired by controlling for subcutaneous fat, ethnic variations appeared. African Americans demonstrated significantly lower visceral fat in relation to levels of subcutaneous abdominal fat compared to Whites.

Like Goran et al (1997) and Bacha et al (2003), Weinsier et al (2001) reported that Whites had significantly more visceral fat than Blacks. This

was demonstrated in their study of 23 White and 23 Black premenopausal women (between the ages of 20 and 46),matched for percent body fat, fat free mass (FFM), total fat mass (FM) and trunk FM.

The literature shows that ethnic differences in fat distribution are consistent across a broad age range and both sexes, when White and Black (mostly Afro American) populations are compared (Bacha et al, 2003; Goran et al, 1997). There is also a considerable amount of research that has investigated fat distribution in Asians, particularly on southern Asians. This is because unlike Black individuals, Southern Asians are known to be predisposed to store more intra-abdominal fat compared to Caucasians (McKeigue et al, 1989) and as a result demonstrate increased metabolic and cardiac risk factors.

He et al (2002) highlighted sex and race differences in fat distribution among Asian (Korean and Chinese), African American and Caucasian prepubertal children. One-hundred and seventy-six girls and 182 boys were examined, aged from 5-12 years. Asian females demonstrated greater relative truncal fat mass than the other groups although the Asian boys had less extremity fat, measured by dual-energy X-ray absorptiometry (DXA), than Caucasians. Wang et al (1994) report that adult Asians demonstrate greater proportions of upper body subcutaneous fat, and that the magnitude is greater in females. In light of this, He et al (2002) concluded that the greater Asian vs Caucasian differences in fat distribution of females compared to males were evident in prepubertal

children as well as adults. From their findings, the authors highlight the importance of sex and race specific interpretations of body composition measurements for validity and to accurately ascertain health risk.

Chandalia et al (1999) examined the relationship of adipose deposition and metabolic abnormalities in Southern Asian men. They attempted to evaluate whether Asian Indians were more insulin resistant than Caucasians and to define the role of generalised and truncal obesity (subcutaneous and visceral fat stored around the abdominal region). They measured height and weight, waist to hip ratio, skinfold thickness at nine anatomical sites, body composition by HW, insulin resistance, glucose disposal and plasma insulin levels in 23 Caucasians of European ancestry and 21 Asian Indians from the Indian subcontinent, temporarily living in the United States. They found that Asian Indians had greater amounts of truncal fat measured by truncal skinfold thickness and higher ratios of truncal to peripheral skinfold thickness. This supports the notion that there is variation in the way that different ethnic groups store fat.

Chandalia et al (1999) reported no significant difference in waist to hip ratio between Asian Indians and Caucasians. This is an interesting finding as a tendency of Asian Indians to store truncal fat is often reflected in reports of increased truncal skinfold thickness and increased waist to hip ratio compared to other ethnic groups (McKeigue et al, 1991; Singh et al, 1995). Chandalia et al (1999) found significantly lower levels of insulin sensitivity in the Asian Indian group, however, this was reported

regardless of the level of total body fat. Therefore a lower insulin sensitivity could not be related to level of body fat or its deposition site. Furthermore, deposition was only determined by truncal skinfold thickness and waist to hip ratio; the use of CT or MRI would allow for the actual quantification of visceral fat. This would provide a better understanding of the relationship between internal fat storage and the prevalence of diabetes and cardiovascular risk factors.

Visceral fat was quantified by Raji et al (2001) who examined the fat distribution of 12 Asian Indians and 12 Caucasians of European ancestry between the ages of 20 and 65 years, living in the USA. Lipid profile, insulin action and anthropometric dimensions including body composition were measured. To quantify internal fat they used CT scans at lumbar 2-3 level and lumbar 3-4 level. Like Chandalia et al (1999), their data show that although matched for BMI age and gender, Asian Indians demonstrated higher abdominal and visceral fat compared to Caucasians. They noticed subcutaneous fat was also higher in Asian Indians compared to Caucasian participants, suggesting Asian Indians had increased generalised central obesity. This increased generalised central obesity was linked to increased insulin resistance and cardiovascular risk compared to Caucasians. The authors found no significant difference between waist to hip ratio of Asian Indians and Caucasians. They put their findings down to alterations in body fat distribution, although they did not find greater risk from any particular compartment of fat. Neither Chandalia et al (1999) or Raji et al (2001) could specifically link visceral fat to

cardiovascular risk or diabetes in Asian men, but they demonstrate a clear link between generalised central obesity and factors associated with cardiovascular disease and diabetes.

Valsamakis et al (2004) also quantified internal fat using MRI to investigate simple anthropometric measures of obesity and their ability to predict visceral fat and the metabolic syndrome. MRI single slice abdominal scans were compared to: waist circumference, BMI, waist to height ratio, waist to hip ratio and sagital abdominal diameter (participant in a supine position, the maximum diameter of the abdomen on the sagital plane) in 83 males (46 diabetics [31 Caucasian, 15 Asian Indian], and 37 non-diabetics [25 Caucasian, and 12 Asian Indian]). They also measured fasting plasma glucose levels and lipid profile as parameters associated with the metabolic syndrome. They reported, irrespective of diabetes and ethnicity, that overall waist circumference was the best predictor of visceral fat mass. However, in the non-diabetic group, age was the best predictor and waist circumference showed a non significant trend. In the diabetic group waist circumference was the best predictor of visceral fat, suggesting those who were diabetic demonstrated increased generalised abdominal (subcutaneous and visceral) obesity. In terms of metabolic syndrome, sagital diameter, waist circumference and age were the best predictors with sagital diameter being the strongest predictor. As sagital diameter includes visceral and subcutaneous fat, the specific compartment making the strongest contribution to metabolic syndrome is unknown and requires evaluation. Further results showed that there was no significant

difference between visceral fat levels between diabetic and non-diabetic groups, other than a trend towards an association of increased visceral fat in the diabetic group. This suggests that visceral fat may not be the primary contributor to diabetes and that centralised subcutaneous fat could play a strong role in the development of diabetes. It was also reported that waist to hip ratio and BMI did not significantly predict visceral fat or metabolic syndrome.

Forouhi et al (1999) investigated how adiposity causes abnormalities at a biochemical level by looking at the relationship between central obesity, insulin sensitivity and muscle cell lipid content in 20 European and 20 South Asian males. They measured anthropometric dimensions, percent body fat and visceral abdominal fat by (DXA) and intromyocellular lipid content (IMCL) by proton magnetic resonance spectroscopy (MRS), and insulin sensitivity. Higher IMCL content was found in South Asians compared to Europeans, but there was no significant relationship to insulin sensitivity in the presence of obesity. Their findings go against the hypothesis that raised IMCL content is associated with insulin sensitivity in South Asians. They reported a positive correlation between IMCL and overall obesity and central obesity and an inverse correlation between IMCL and insulin sensitivity in Europeans. However, in South Asians it was found that IMCL was not significantly correlated with any measured variable, although insulin sensitivity was associated with plasma fasting triglyceride and waist to hip ratio. Forouhi et al (1999) postulate that an

increased supply of triglycerides from plasma, consistent with their data, could contribute to insulin resistance.

It is evident that although matched for BMI, South Asians demonstrate a lower sensitivity to insulin than Europeans generally (Forouhi et al, 1999). This is seen at BMI levels within the healthy range (18-25 kg/m^2) supporting the understanding that Southern Asians are predisposed to a reduced insulin sensitivity regardless of BMI classified obesity. However, Forouhi et al (1999) do not explain the relationship between BMI and body composition. It is possible, that as with other studies using Southern Asian participants, a healthy BMI is associated with a percentage body fat that classifies the participant as overweight or obese, and that the decreased insulin sensitivity is due to raised adiposity which is poorly defined by BMI. This is supported when the mean values of Forouhi et al (1999) measurements are examined. Europeans and Southern Asians both demonstrate almost identical mean BMI values (26.4 ± 0.8kg/m^2 and 26.3 ± 0.6kg/m^2, respectively), however, DXA-measured %BF is 22.5 ± 1.7% in Europeans but 26.8 ± 1.3 in Southern Asians. This highlights the inaccuracy of BMI for identifying obesity risk when used on different ethnic groups. This suggests that BMI is not comparable across different ethnic populations and that Southern Asians seem to have greater proportions of body fat when matched for BMI to Caucasians.

Increased central obesity was linked to glucose intolerance, hyperinsulinemia, low HDL cholesterol and high fasting triglyceride levels in Southern Asians compared to Europeans (McKeigue et al, 1991). They postulate that this could be due to central obesity being linked with a failure of insulin to suppress the release of non-esterised fatty acids (NEFA) from intraabdominal fat cells, as internally stored fat cells are less sensitive to the antilipolytic action of insulin (Yki-Jarvinen & Taskinen, 1988). McKeigue et al (1991) highlight that this failure would increase the synthesis of LDL triglyceride at the liver, increasing the circulating volume of triglyceride in the blood. McKeigue et al (1991) state that increased triglyceride production due to increased central obesity and visceral fat seems to mediate other effects on lipid metabolism. They state this could be directly linked to atherogenesis, which is associated with cardiovascular disease. These findings highlight the importance of data that can quantify non-esterised fatty acid levels. It is important to note that central obesity was determined by waist to hip ratio and skinfold analysis. Waist to hip ratio has been linked to visceral fat level by Caprio et al (1993) and Banerji et al (1993). However, Chandalia et al (1999) found no relationship between waist to hip ratio and visceral fat, thus making it difficult to determine, although centrally stored, whether abdominal subcutaneous or abdominal visceral fat is predisposing patients to health risk.

More recently, Abate et al (2004) quantified NEFA levels, when they investigated adipose tissue metabolites and insulin resistance in non

diabetic Asian men (79 Asian Indian and 61 Caucasian men). They measured anthropometry and blood parameters after a glucose tolerance test. Hydrostatic weighing was used to calculate body composition. The main findings were: plasma NEFA are raised and insulin mediated plasma NEFA suppression is impaired in Asian Indian men compared to Caucasian. Plasma leptin concentrations were higher in the Asian group and plasma adiponectin concentrations were lower in Asian Indians (adiponectin regulates plasma glucose levels and fatty acid catabolism; Diez & Inglesias, 2003). These findings support the work of Forouhi et al (1999) and Mckeigue et al (1991) who also found Asian Indian men had greater levels of plasma triglyceride, which they propose could cause insulin resistance. Abate et al (2004) reported that adiponectin was most strongly correlated with raised truncal skinfold thickness in both groups. As Asian Indians had higher truncal skinfold thickness they assessed the differences between the two groups after adjusting for body fat content, truncal skinfold thickness and waist circumference. This revealed that plasma adiponectin levels were significantly lower in Asian Indians independent of obesity. They postulate that, in the absence of obesity, a raised NEFA and leptin concentration and a decrease in adiponectin concentrations in Asian Indians in general, seems to represent a genetic susceptibility to insulin resistance. This supports Forouhi et al (1999) and Chandalia et al (1999) who also found a greater prevalence of cardiovascular disease risk factors in Asian Indians regardless of obesity level. Abate et al (2004) suggest, that although seemingly genetic, these

abnormalities are likely to be accentuated by increasing obesity levels observed in Asian Indians.

The literature clearly identifies a link between obesity and health risk. In particular central obesity, increasing the prevalence of metabolic abnormalities that are thought to promote type II diabetes and cardiovascular disease. There are obvious ethnic differences in obesity and the disorders associated with it. Southern Asians seem to carry more central fat than Caucasians, although there is conflicting evidence of whether it is stored subcutaneously or viscerally. Afro-Caribbeans seem to store less centralised fat than Caucasians. These differences are seen across a broad range of ages from very young children to adults, up to the age of 60 years. However, obesity level does not consistently correlate with health risk, as a number of studies have reported Asian Indians to be insulin resistant and more prone to diabetes regardless of obesity level or the deposition of their body fat. This suggests that Asian Indians have a genetic susceptibility towards type II diabetes. Although there is some conflicting evidence as to relation of obesity and health risk, there is certainly an association between centrally stored body fat and health risk, particularly in Asian Indians. Thus making Asian Indians a high risk population, predominantly due to their association with type II diabetes. This knowledge highlights the importance of accurate identification of obesity in order to identify potential health risk, and in particular, high risk Asian Indians.

Identifying obesity

Accurate identification of obesity and the associated potential health risk is vital in order to treat and reduce the ill effects obesity has to health. Body mass index (BMI) (mass[kg]/height[m]^2) is currently the most common diagnostic tool used in research and clinical settings to classify overweight (25-29.9 kg/m^2); obese (>30 kg/m^2) and underweight (<18.5 kg/m^2), as defined by the World Health Organisation (WHO, 1998). BMI has been identified as a significant predictor of type II diabetes and cardiovascular disease (Janssen et al, 2002) because of this, and its simplicity, BMI is used in many prospective and population based studies to identify individuals at risk. However, BMI is limited as it does not account for the composition of a person's body weight. A very lean individual may be misclassified as overweight or obese according to BMI tables, when in reality they are quite the opposite. This is due to the greater mass of muscle tissue causing the individual to appear heavier for their height, but not due to excessive adiposity. This is because lean tissue has a higher density (1.100g/ml) than fat tissue (0.9007g/ml) (Schutte et al, 1984). Therefore, it is important to determine body composition, the percentage of fat mass and fat free mass that contributes to total body mass in order to truly identify if an individual has a healthy body fat level. BMI is also limited by factors such as ethnicity, age, frame size and body build that affect the relationship of BMI and percent body fat. This was highlighted by Banerji et al (1999) in their study of regional adiposity and cardiovascular risk in 20 healthy Asian Indian male volunteers with no known history of diabetes. They found a mean body mass index of 24 ± 2.54kg/m^2,

classifying the participants as neither obese or predisposing to diabetes but when percent body fat by computerised tomography was measured, mean total body fat was 33 ± 7 %BF, placing them as significantly obese. This highlights the importance of ethnicity specific cut points in order to accurately identify health risk for different ethnic groups (Mckeigue et al, 1992; Seidell, 2000). Because of the misleading nature of BMI, a major aim of research in the field of applied body composition assessment is to develop accurate field methods to estimate body fat.

Body Composition

Body composition measurements are used to quantify proportions of a persons body fat, expressed as a percentage of their body weight. There are a number of methods based on different assumptions that vary in terms of validity. Most models used to estimate body composition (2, 3 and 4 compartment models) are based on the same initial principles that build upon each other as the techniques become increasingly advanced and valid. The most recent advances in body composition use new technology, such as magnetic resonance imaging (MRI), dual-energy X-ray absorptiometry (DXA) and computerised tomography (CT), that, although very costly, are more accurate than the more basic models of body composition.

Two-compartment model

The most simple and common method of estimating body composition is the two-compartment model (2C), which separates the body into two

compartments, a fat compartment and a fat free compartment, expressed as %FM and %FFM, respectively. FM is the absolute amount of body fat consisting of all extractable lipids from adipose and other tissues; FFM consists of all remaining chemicals and tissues including water, muscle, bone, connective tissues and internal organs (Heyward & Wagner, 2004). The 2C is based on the measurement of total body density (Db) and assumes the density of FM and FFM to be constant, using standardised algorithms based on these densities to estimate %BF. Keys and Brozek (1953) developed a 2C equation based on a reference cadaver that consisted of 14% BF and assumed the density of fat was 0.9478 g/cc. Ten years later this equation was revised by Brozek et al (1963) using a reference cadaver with an assumed density of 1.064 g/cc and 15% body fat which produced a more accurate fat density of 0.9007 g/cc. From this model, any variation in measured Db to the reference body density (1.064g/cc) is assumed to be due to variations in adipose tissue. This equation, Percent body fat = (4.57/Db – 4.142) x 100, has been used widely to obtain 2C estimates of body composition. Siri (1956) also developed a 2C equation to convert Db to percent bodyfat. It uses constants different to that of the Brozek et al (1963) equation. The Siri (1956) equation, Percent body fat = (4.95/Db – 4.50) x 100, assumes that any variation in measured Db from that of the reference body is due to variation in triglyceride content instead of adipose tissue. When the two equations are compared they produce nearly identical body fat estimates. For example, if measured Db is 1.0500 g/cc, the estimated body fat percentage from the Siri (1956) and Brozek et al (1963) equations will be

21.4% and 21.0% respectively (Heyward & Wagner, 2004). However, the Brozek et al (1963) equation is deemed most appropriate when measuring those who are very adipose or very lean, due to the greater degree of bias produced by the Siri equation in individuals expressing these characteristics.

Three compartment model

Taking into account interindividual variation of the hydration of the fat free body (FFB), Siri, (1961) developed a three compartment (3C) model. It divides the body into three compartments: fat, water, and solids (mineral and protein fractions of the FFB combined) assuming a constant density for the protein to mineral ratio. The 3C model measures the hydration of the FFB, so does not need to assume it (Heyward & Wagner, 2004). Thus, it is understood that the 3C model may produce more accurate estimates of percent body fat for individuals or population subgroups whose hydration of the FFB is not consistent with that assumed (73.8%) by the 2C model (Segal et al, 1987), such as obese adults or children. Later, Lohman (1986) devised a 3C model that accounts for mineral content of the FFB, dividing the body into fat, mineral, and protein + water fractions. It assumes a constant density of 1.046 g/cc for the protein and water fraction. The relevant measurements for this model are Db by densitometry and total body mineral (TBM) by dual-energy X-ray absorptiometry (DXA), TBM consists of osseous and non-osseous mineral. The Lohman (1986) 3C model produces more accurate estimates of percent body fat compared to the 2C model for individuals whose TBM

varies from the assumed 6.8%FFB, such as African American men (Schutte et al, 1984; Wagner & Heyward, 2001). DXA also uses a 3C tissue level model which divides the body into bone- free lean tissue mass (LTM), FM and bone. The 3C DXA model uses two separate 2C model equations (Ellis, 2000). The first set of equations divides the body into bone and soft tissue mass (STM); STM is fat + LTM. The second set of equations separates the STM into fat and lean tissue. Through this model DXA is capable of separating the body into bone and STM. Lohman et al (2000) concluded that DXA estimated percent body fat within 1% to 3% of multicomponent molecular model estimates.

Four compartment model

The four compartment (4C) model divides the body into fat, water, mineral and protein, thus, removing the need to assume proportions of any of these constituents in the body. Reference methods are required to measure Db, TBW and total body bone mineral (TBBM). The 4C model has greater accuracy than the 2C model when estimating percent body fat (Heymsfield et al, 1996). However, the 4C model requires the measurement of more variables than the 2C model. The cumulative errors associated with multiple variable measurement has been assessed to investigate whether these errors offset the improved accuracy in estimating body composition. This was done by Freidle et al (1992) who compared the measurement error of the 2C model with those of the 4C model. They reported errors of ±1.0 %BF and ± 1.1 %BF, respectively, for the 2C and 4C models leading to the conclusion that the error produced by

the four separate variable measurements of the 4C model does not offset its greater accuracy compared to the 2C model.

Six compartment model

The six compartment (6C) atomic model utilises the direct analysis of chemical composition of the body *in vivo*. The total body content of the major elements (i.e. calcium, sodium, chloride, phosphorous, nitrogen, hydrogen, carbon and oxygen) can be measured using neutron activation analysis (NAA). Wang et al (1998) have developed a 6C model which divides the body into water + nitrogen + calcium + potassium +sodium + chloride. The accuracy of the 6C atomic model is high enough to provide criterion measures for evaluating other reference methods and models. However, the lack of NAA facilities, very high expense, and high participant radiation exposure drastically limits the use of such a model.

Computerised tomography and magnetic resonance imaging

Other highly advanced laboratory techniques are computed tomography (CT) and magnetic resonance imaging (MRI). These techniques take physical images below the surface of the skin that can be used to quantify proportions of fat throughout the body. As a result they do not need to assess body composition at a molecular level. CT measures attenuations of X-ray beams as they pass through the body. Varying densities of the underlying tissues produce attenuation differences in the X-ray beams that create a computer generated image of the area scanned. These images allow for the separate recognition of lean tissue, bone and adipose tissue.

Although highly accurate, this technique is limited by high cost and low availability. Radiation from X-rays often limits CT scans to regional assessment rather than whole body. This is because exposure to such radiation is harmful and prolonged exposure may be deemed un-ethical. MRI, however, does not use ionizing radiation; It uses an external magnetic field and then a pulsed radio signal frequency that is passed across the body. When the radio waves cease, the radio signals are emitted back from the tissues and used to create a computer generated image. These techniques allow for tissue level analysis of body composition, they are the best way of accurately separating adipose tissue into subcutaneous and visceral fat. MRI is particularly advantageous as it can be used for accurately analysing total whole body fat (Kullberg et al, 2009) including visceral (Abate et al, 1994; Thomas et al, 1998) due to the fact that it emits no radiation (Heyward & Wagner 2004). However, both CT and MRI are both very costly and medical based systems such as these have limited general access.

Thomas et al (1998) used whole body MRI to assess total body fat and concluded it was a reliable and non-biased measure of body fat content (subcutaneous and visceral) in a varying body shapes and sizes. Single-slice MRI was found to be unable to predict the large variation of individual visceral fat content. Later, Thomas and Bell (2003), compared single-slice MRI and multi-slice MRI (L2-L3 + L4-L5) for the measurement of intra abdominal adipose tissue content. Fifty-nine healthy females were examined, 17 were included in a 6 month exercise intervention to assess

measurement of change in visceral fat content. Although single-slice appeared to be suitable for measuring change in visceral fat, it was deemed to be inconsistent when quantifying total visceral fat content. Multi-slice, however, was shown to provide precise determination of total visceral fat content.

Unfortunately the limitations associated with MRI and CT such as hazards to health (X-rays), high expense and limited availability are also common among the more accurate reference methods utilised in order to estimate body composition via the 3C, 4C or 6C. Although the 3C and 4C models are used more regularly than the very rare 6C model, they still require the use of expensive techniques that have relatively limited availability. Some such as DXA, expose participants to low levels of radiation which may make participants wary of participating, especially over a longitudinal study involving many repeated exposures to radiation. This also limits studies to regional scanning such as a single slice of the abdomen, rather than the whole body. This highlights the usefulness of research that aims to validate 2C model techniques in order to improve their accuracy.

Advantages of the two-compartment model

The main advantage of the 2C model is the relatively low cost of the associated devices and techniques compared to the most current reference devices that are deemed most accurate. Another advantage is the small size of many of the 2C model devices, they are very mobile and can be taken out into the field. Further to this, is their ease of use and the

short time period taken to collect and interpret data from the majority of 2C devices. Most 2C devices take a few minutes to prepare and a matter of seconds to produce meaningful data. A number of more advanced techniques such as MRI, although very accurate, can take much longer to prepare, measure and produce meaningful data, some techniques require raw data to be analysed and interpreted, prolonging the data collection process.

Researchers (Duz et al, 2009; Elberg et al, 2004; Wagner et al, 1999) have assessed the validity of the 2C model and other field methods using such devices as hydrostatic weighing (HW), air displacement plethysmography (ADP), bioelectrical impedance analysis (BIA), and skinfold thickness analysis (SKF) to those deemed more accurate, such as the 3 and 4 compartment models and more advanced techniques such like DXA.

ADP (measured by the BOD POD) has been compared to HW in the past in order to determine its accuracy against the 2C “gold standard” for estimating body composition (Lohman, 1981). Wagner et al (1999) identified that the BOD POD was an attractive method due to its simplicity to operate in a short time period compared to HW. However, significant overestimation of percent body fat lead to recommendations that more cross-validation research is required before ADP can be used in place of HW. Later, Wagner et al (2000) stated that ADP method could potentially replace HW due to its more accommodating procedures, thus reducing subject error. Percentage body fat for both methods were subsequently

compared with that obtained by DXA to gain a reference measure (Wagner et al, 2000). ADP was found to significantly overestimate percent body fat by 1.73%, compared to DXA, HW only varied by 0.25 %BF. This suggests that HW produces a more accurate estimate of %BF than ADP. It could be suggested that the close relationship between HW and DXA could allow for HW to be a less costly substitute to DXA.

ADP was also compared to DXA to validate its ability to estimate body fat change over time by Elberg et al (2004). Initial findings found ADP was not significantly different to DXA. However, further statistical analysis revealed a significant magnitude bias of ADP, suggesting ADP could not be a useful substitute for DXA. Elberg et al (2004) also compared BIA and tricep skinfold thickness (TSF) to the criterion. BIA overestimated percentage body fat whilst TSF could not account for more than10% of the variance in DXA. These findings suggest BIA and TSF are not acceptable substitutes for DXA. Although BIA and TSF are much cheaper alternatives to DXA, it is likely the improved accuracy of DXA outweighs the advantages of BIA and TSK (i.e. low cost, availability and ease of use). The data suggest that ADP could be a useful tool for measuring body fat change.

Radley et al (2003) also compared ADP to DXA and like Elberg et al (2004) found a high correlation between the two devices, although further analyses revealed unacceptably high limits of agreement between the two methods. They highlight that the benefits of ADP warrants further investigation into its validity. ADP was found to produce acceptably similar body fat measures when compared to the criterion HW (Moon et al, 2008),

which has been suggested by Wagner et al (1999) as a potential substitute for DXA. Moon et al (2008) also compared SKF to HW and found it was a superior method than ADP for estimating body fat.

Duz et al (2009) compared SKF and BIA to DXA in men and women. Both SKF and BIA underestimated body fat compared to DXA in both sexes. However, BIA produced the closest percent body fat compared to DXA in males but BIA showed a magnitude bias as body fat increased. This suggests that the two methods (BIA and SKF) cannot be used interchangeably and that BIA is a superior estimate of body fat compared to SKF, but should possibly not be used in those who are very obese.

Although the 2C model has less accuracy than the more advanced 3C and 4C models, the devices utilised by the 2C are much less costly and easier to operate. Most 2C model techniques can be used easily in the field for fast evaluation of body composition. These benefits will often outweigh the improved accuracy of reference measures which is shown by their continued use in research. Less portable 2C methods such as ADP and HW tend to be regarded as the most valid when compared to reference measures such as DXA (Elberg et al, 2004 & Wagner et al, 2000).

Disadvantages of the two-compartment model

The principles behind the 2C makes assumptions that if not met will lead to error in the measurement. These are: that the densities of fat mass (FM) and fat free (FFM) mass components are additive and the same for

all individuals; that the proportions of water, mineral and protein making up the FFM or reference body are constant between and within all individuals; and that the only difference between the individual being measured and the reference body is the amount of adipose tissue or body fat. This means that controllable parameters such as hydration between participants must be consistent for valid and reliable data.

Different 2C devices require control of different parameters for valid estimations of body composition. BIA specifically requires participants to be normally hydrated as hydration level impacts on the impedance of the current and the resulting fat free mass estimation. BIA is based on the principles of electrical conductivity, combined with basic assumptions of the geometric shape of the body and of the relationship of impedance (opposition to flow of current) to the volume and length of the conductor (Heyward & Wagner 2004). The body's tissues act as conductors and insulators to electrical current, which will flow through the body taking the route of least resistance. Fat free tissue is composed of water (73%) and electrolytes (Heywood & Wagner, 2004), and is therefore a better conductor than fat, which is anhydrous. It is the impedance of this current which is measured by BIA devices and is directly correlated to the amount of water in the body, known as total body water (TBW). Due to fat free tissue being composed of 73% water, FFM can be predicted from TBW. As a result of these principles, dehydration will result in an overestimation of fat mass.

ADP and HW are both densitometric methods, ADP is based upon air displacement and HW on water displacement. They are both known to be affected by multiple factors. Moisture on the surface of the skin (ADP only), excess body hair, trapped air in hair and loose fitting clothing, and gas in the gastrointestinal tract are all known to impact upon ADP and HW estimates of body composition (Fields et al, 2004).

The equations used to calculate ADP and HW measure body composition require values of thoracic gas volume (TGV) and residual lung volume (RLV) respectively. The BodPod hardware includes a breathing tube accompanied by software that measure TGV, however, this value can also be predicted using equations based on age and height prediction tables of TGV. Predicting this value is quicker than measuring, as measurement requires a difficult breathing technique that some individuals find difficult to perform, it takes no additional time, and although less accurate, is often chosen over measuring in clinical settings. Measuring RLV for the HW method requires advanced techniques such as closed circuit helium dilution, hence the same conflicts arise concerning accuracy over practicality. This is an advanced and costly technique, requiring tester expertise. This value can also be predicted via simpler and less expensive methods, RLV can be predicted from forced vital capacity (FVC) measurements and these can be obtained via highly portable micro spirometers. However, they require participants to maximally exhale preceding a maximal inhalation which can be difficult to perform if not well practiced. As a result individual differences will act upon these predicted

values, potentially adding to the error of the estimations of body composition through ADP and HW (see equations in appendix B).

A major issue brought about by these assumptions is that of a consistent FFM density between all individuals. Research has shown that this assumption is not met when measuring different ethnic populations. This is because different ethnic groups demonstrate different FFM densities (Ellis, 1997; Schutte et al, 1984). This means that the 2C is not comparable between different ethnicities within a sample; more importantly, the 2C is not a valid measure for non White populations. This is due to the reference bodies used by Siri (1956) and Brozek et al (1963), as they examined the cadavers of white individuals for the development of their equations. This means the 2C model is population specific to Caucasians and has led to the development of new equations for different ethnic groups. However, there is very limited research into the validity of the 2C model on Southern Asians and there are no validated correction equations developed for this population. The fact that Southern Asians are a high risk population when it comes to obesity related disorders is a concerning one, again raising the issue of the accuracy of body composition identification and potential health risk in this ethnic group.

Ethnicity and density- Implications for the two compartment model

Schutte et al (1984) identified the issue of varying FFM density in different ethnic groups and how the 2C may be invalid as a result. The authors realised that the increased density of skeletal mass in Blacks compared to

Whites upon whom the 2C algorithms are based could significantly increase the overall FFM density of black participants. This would mean the constant value for FFM density used in the 2C equations are inappropriate and potentially invalid when used on Black participants. Schutte et al (1984) measured Db, TBW and anthropometric dimensions in 19 White and 15 Black males between 18 and 32 years of age. Black and White participants demonstrated similar height, weight and age. The authors compared lean body mass and total body fatness derived from observed density and also TBW. Body composition from densitometry and TBW yielded very similar results in the White participants. However, among the black participants, body composition based on densitometry produced a significantly greater lean body mass and significantly lower percent body fat than those based on TBW. Lean body mass and percent body fat by TBW in the Black participants were nearly identical to those produced by the White participants who were similarly matched for height and weight, suggesting that differing scores of body composition were due to varying body densities between the Black and White participants. They also found little difference between observed and predicted density in White participants. As expected Black participants demonstrated a significantly greater observed density than predicted. Due to the anthropometric similarities between the two groups the predicted densities of both groups were very similar. The Schutte et al (1984) findings were consistent with the hypothesis that Black individuals have a denser lean body mass than whites. These differences led to overestimation of the lean body mass and an underestimation of percent body fat. The authors

calculated that the Black participants demonstrated a FFM density of 1.113 g/ml rather than 1.100 g/ml based on White cadaver data. In order for this variation in density, Black participants must have a 36% greater bone mineral density if the differences are entirely down to a denser skeletal mass (as the authors first proposed). They suggests that as a 36% greater bone mineral density falls outside of 10-20% range observed *in vitro* skeletal studies (Merz et al, 1956; Seale, 1959), that a greater mineral and or protein content must also be contributing to a greater lean body mass density observed in Black individuals. These finding led the authors to develop a 2C formula to convert body density into percent body fat for Black men: Percent body fat = [(4.374/Db) – 3.928] x 100, based on their calculations that Black men have a fat free body density of 1.10570 g/ml. This new formula is now generally used for estimating percent body fat from Db in Black men to the present day. However, its validity and generalisability are not fully known. The formula was not cross validated until 2000 by Wagner and Heywood (2000). TBW was used as the reference method of estimating percent body fat rather than a more precise multicomponent model. Furthermore, the data used to create the conversion formula were from a small, homogeneous sample (n = 15) of young, Black college students aged 18-32 years, and cannot be generalised to a wider population. It was Wagner and Heyward (2000) who first cross validated the Schutte et al (1984) equation using a four compartment model as the criterion. The four compartment model does not need to assume the density of the fat free body and, therefore, is unaffected by varying FFM density observed in different ethnic

populations. They also examined the validity of the Siri (1956) and Brozek et al (1963) 2C equations for estimating body composition on their sample of Black males (n = 30) aged between 19 and 45 years. They found that the Siri (1956) and Brozek et al (1963) equations significantly ($P \leq 0.01$) and consistently underestimated percent body fat in Black males by 1.94 and 1.75%, respectively. They also found under close inspection of residual scores that 87 and 90% of the sample were underestimated, respectively. Furthermore, the Schutte et al (1984) equation significantly ($P \leq 0.01$) and systematically (87% of sample) overestimated percent body fat by 1.28%, leading to the authors developing a new 2C formula for body composition estimation in Black males. Due to the greater sample size and the fact that it was based on a multicompartment model, the authors recommend using their conversion formula and not that of Schutte et al (1984) when converting Db to percent body fat in this ethnic group.

These findings are supported by Ellis (1997), who like Schutte et al (1984), found that BMC and LTM were significantly higher in Black than in White males, aged of 3 to 18 years. This would mean that the Black males had a higher FFM density than the White males. This could suggest that Black males demonstrate an increased FFM density from birth or a very young age. This would mean that ethnic differences in FFM density and their implications on 2C measured body composition in young individuals may also benefit from re-consideration. Interestingly no significant difference was identified in BMC and LTM between White and Hispanic participants,

suggesting that body density will not affect the validity of 2C measured body composition of young, healthy, Hispanic male individuals.

It is evident that the increased density of this Black individuals warrants the use of separate formulae for the estimation of body composition on Black individuals. It also highlights the need for further investigation into the FFM density of other non White ethnic groups and its implications for body composition estimation and in turn accurate health assessment, especially in Asian Indians, who have been highlighted as a high risk population. It is very important that research investigates this population further. Although there are many different nationalities and sub-groups within the Asian population, the majority of research has been conducted on Japanese and Chinese participants. As such, there are few validation studies on the accuracy of 2C model measured estimates of body fat in Asian Indians. A better understanding of the use of the low cost and easy to administer 2C model in Asian Indians, that can be applied to the field, will greatly improve our ability to assess obesity and potential related health risks in this ethnic group.

3. Methodology

In order to infer validity of measurement, it is important to determine the reliability of the devices tested. Therefore, reliability data was collected before main testing started. The MRI scanner (Philips Achieva 1.5T, Heidoven, The Netherlands, criterion measure) and Harpenden skinfold calipers (CMS Instruments, London, UK) were measured (by ISAK trained researchers) at the Hammersmith hospital through a collaborative research project at Imperial College London. Therefore, these devices were regularly tested for reliability and calibrated by technicians at the Hammersmith hospital. As a result it was not required to include them in the current reliability testing. Reliability testing was conducted at the University of Bedfordshire, at the Bedford Sport and Exercise Science Laboratory, Polhill Campus. For all testing protocols, see Appendix A.

Reliability testing Procedure

Twelve male subjects aged 19-26 years were recruited from the student population of the University of Bedfordshire. Inclusion criteria were that the participant was male and a university student aged between 18 and 55 years.

After reading informed consent with accompanying information sheets the participants agreed to take part by signing the consent document and a PAR-Q questionnaire to confirm their state of health (see Appendix B). Participants were required to provide their own tight fitting clothing such as swimming trunks or Lycra shorts. Prior to testing participant were to have

required to be fasted and to have refrained from physical activity for 4 hours. Participants were reminded by telephone call of their appointment in the laboratory one day prior to testing. They arrived at the sports science laboratory of the University of Bedfordshire (Polhill campus) in pairs at a pre-arranged time. Upon arrival they were instructed to use the toilet to void the bladder and bowel in order to reduce a false body volume effect from urine, feces and gastrointestinal gas when measured using ADP and HW. At this point they were also instructed to change into tight fitting clothing in the privacy of a changing room. A robe was provided to wear between testing or during tests that did not require minimal clothing. Some participants preferred to wear their own t-shirt instead of a robe.

Each participant had three repeat measures of body composition on all devices. All measurements were taken on the same day for each 2C model device. The apparatus tested were: Tanita® Segmental body composition analyser, BC-418, Tanita, Holland; the Bodystat® 1500; Bodystat, Douglas, United Kingdom (Bodystat [BIA]), the BOD POD®, Life Measurements Instruments, Concord, CA, U.S.A. (ADP); and the University of Bedfordshire underwater weighing tank (HW). For protocol of all devices, see Appendix A.

Intraclass correlation coefficient and coefficient of variation between repeated measures were calculated using Hopkins (2000) to determine if error between repeated measurements was too high to be reliable. Its vital that measurement error is not too large to infer a correction equation for Southern Asian Indians.

Main data collection procedures

Acceptance criteria for participation were: participants must be male, aged between 18 and 55 years, healthy but sedentary and to be classified as Asian Indian, (all four grandparents must be Asian Indians). All four grandparents must be Caucasian to classify as Caucasian.

Twenty-one males participated in the study, comprised of 11 Caucasians and 10 Asian Indians aged between 21 and 51 years of age. Participants were made up of staff and students of the University of Bedfordshire. Descriptive characteristics of the participants are shown in Table 1. Caucasians were older than Asian Indians with a greater range of ages (33.36 ± 11.67 and 26.80 ± 4.61 years, respectively). Caucasians were slightly taller than Asians Indians (1.79 ± 0.08 and 1.70 ± 0.04 meters, respectively). Caucasians were considerably heavier than Asians (83.85 ± 9.30 and 73.21 kg, respectively) and had a greater BMI than Asian Indians. However, Asian Indians demonstrated higher percent body fat assessed by MRI compared to Caucasians.

Table 1. Descriptive characteristics of all participants (mean, standard deviation and range)

	All participants (n=21)			Caucasian (n=11)			Asian (n=10)		
	M	**SD**	**range**	**M**	**SD**	**range**	**m**	**SD**	**range**
Age (years)	30.24	9.94	31.00	33.36	11.67	30.00	26.80	4.61	18.00
Height (m)	1.75	0.08	0.25	1.79	0.08	0.25	1.70	0.04	0.13
Weight (kg)	78.78	11.15	49.80	83.85	9.30	34.20	73.21	10.70	37.10
BMI (kg m²)	25.81	25.81	13.19	26.19	3.34	11.13	25.38	3.85	13.17
BF% (MRI)	23.24	8.73	31.24	17.18	6.49	21.00	29.91	5.31	16.69

(m= mean, SD=standard deviation).

Two-compartment (2C) model methods

Prior to participants arriving at the laboratory and for participant preparation i.e. clothing and use of the toilet; the same procedures were followed as explained above for reliability testing. Participants had body composition measured by 2C model methods on one day and were scanned by MRI on a separate day. 2C model measurements and MRI were taken within one week of each other. The number of participants booked to be MRI scanned determined the number of participants measured using the 2C model each week, as only one day per week (Tuesday) was allocated to MRI scanning. As all 2C model methods were tested on the same day, it was important to ensure HW (underwater weighing tank) was the last technique used as excess moisture on the skin can affect the accuracy of the other measurements. The order was as follows: BIA (Tanita and Bodystat), ADP (BodPod) and HW (underwater weighing tank). Testing protocol can be found in Appendix A.

Statistical analysis

A three-pronged approach was taken in order to assess the validity of the five predictor variables compared to the criterion. The difference between MRI, ADP, BIA HW and SKF percent fat estimates was examined using mixed measures ANOVA to assess the differences of the mean percent body fat scores both within and between Caucasian and Asian groups. Correlation coefficients and stepwise multiple linear regression analysis were used to assess the linear relationship between the criterion and predictor variables and agreement between body composition estimates was examined by calculating the 95% limits of agreement as explained by Bland and Altman (1986).

The use of multiple regression allows for the prediction of MRI body fat from one or more predictor variables. Potential bias between MRI percent body fat and the predictor variables was assessed using residual plots. For all analysis the alpha level set for statistical significance was $P<0.05$, using Statistical Package for Social Sciences (SPSS) for Windows (version 16.0).

A mixed measures ANOVA is a parametric test which makes the following assumptions of the data, that if not met increases the chance of committing a type I error (rejecting the null hypothesis when it is true): The sample data is normally distributed; samples have equal variance; the dependent variable is measured on an interval scale and that there is sphericity of the within groups comparisons. To test for normally distributed data amongst a small sample size, the Shapiro-Wilks statistic

can be examined (see Appendix E). This tests the null hypothesis that the data is normally distributed. Shapiro-Wilks shows that all body composition variables, other than Caucasian Bodystat (BIA), assume normal distribution ($P > 0.05$). As Caucasian Bodystat (BIA) demonstrates non-normal distribution ($P = 0.038$) the skewness and kurtosis statistics can be referred to (see appendix C [Caucasian] and C-1 [Asian Indian]). This variable is more skewed (0.480) and kurtotic (2.462) than the other body composition variables suggesting it may have non-normally distributed data and that a parametric test such as ANOVA should not be used upon it, due to the increased chance of committing a type I error. However, according to Vickers (2005) the usefulness of ANOVA when data is not normally distributed can outweigh the use of non-parametric alternatives. The Levene's statistic (see appendix E) tests the null hypothesis that there is no significant difference between the variance of the two ethnic groups. All variables included in the mixed measures ANOVA, show a significance greater than 0.05, thus, equal variance is assumed. All data is on an interval scale, e.g. the difference between each unit of measurement is always equal (the difference between 1% and 2% body fat is equal to the difference between 21% and 22%). The Sphericity assumption assumes the variance of the pairs of scores, contrasted within groups, is not significantly different Mauchly's test of sphericity (see appendix B) shows the null can be accepted ($P = 0.152$) and sphericity is assumed.

Further more, multiple regression makes the following assumptions: the relationships between the predictor variables and the criterion variable

should be linear; the residual scores produced by the regression model should be normally distributed; the residual variance show homogeneity (should be constant) and the residuals associated with one observation are not correlated with the errors of any other observation, known as independence. The assumption of linearity was represented in table 3, that showing positive relationships of all the variable used in the regression equation. To see if the data is normally distributed, the normality curve of the residual scores of the dependent variable against the predictor variable and the P-P plot of the standardised residuals can be observed (appendix F). The data is not radically different from the normality curve for both Caucasian and Asian Indian regression analyses although the histograms show some degree of positive kurtosis. The P-P plots for both groups, particularly Asian Indians, show the data points lie close to the normality line although there is winding around the normality line indicative of the kurtosis observed in the normality curve of the residual scores. The residual plots (appendix F) appear to display random distribution throughout the xy space indicates the variance is homogenous. The assumption of independence seems to be met as the collinearity statistics (Appendix F) shows that the tolerance and VIF values of the variable included in the regression models are well within acceptable ranges.

The regression equation is calculated from the following formula, based on the slope and intercept of the regression line:

$Y= a_y + (b_y)(x)$

Where *a* is the Y-intercept and *b* is the slope of the line, x is the value of the predictor variable. This predicts Y which is the predicted value of the criterion variable.

4. RESULTS

Mean body composition values

Mean percent body fat for all six body composition devices are displayed in Table 2. Repeated measures ANOVA reveals between groups comparisons show all methods produced higher percent body fat scores in Asians than Caucasians. This was shown by a significant Caucasian versus Asian mean difference between all methods other than BIA-bodystat (15.98 and 19.01% body fat, respectively). The greatest difference was between MRI measured percent body fat, 17.18% (Caucasian) and 30.29% (Asian Indian).

Table 2. Mean body fat percentage measured by skinfolds, bioelectrical impedance, air displacement plethysmography, hydrostatic weighing and magnetic resonance imaging displaying between groups difference [means and (standard error)].

	Caucasian (n=11)	**Asian Indian (n=7)**	**Between groups difference**	
Skinfolds	19.57 (1.48)	26.53 (1.86)	6.95 (2.37)*	P= 0.010
BIA-Bodystat	15.98 (1.23)	19.01(1.54)	3.03 (2.37)	P= 0.144
BIA-Tanita	16.08 (1.51)	22.00 (1.89)	5.92 (2.42)*	P= 0.026
ADP	20.36 (1.97)	30.09 (2.47)	9.73 (3.16)*	P= 0.007
HW	18.91 (1.81)	27.64 (2.27)	8.74 (2.91)*	P= 0.008
MRI	17.18 (1.88)	30.29 (2.35)	13.11 (3.01)*	P= 0.000

Correlations

Table 3 shows that all 2C methods, other than Bodystat (BIA), have a significant, positive linear relationship with MRI in the Caucasian group. ADP has the strongest positive linear relationship ($r = 0.878$, $P \leq 0.0001$) with MRI. In the Asian group, all 2C methods other than Tanita (BIA), have a significant, positive linear relationship. It is important to note that only seven participants were tested using the Tanita (BIA) device in the Asian Indian group. The strongest positive significant linear relationship with MRI was demonstrated by the comparison of tricep skinfold thickness and that of skinfold analyses ($r = 0.852$, $P = 0.004$ and $r = 0.821$, $P= 0.007$, respectively). In the Asian Indian group, skinfold thickness measurements (bicep SKF, tricep SKF and subscapular SKF) had stronger correlations with MRI percent body fat ($r > 0.807$) compared to the Caucasian group ($r < 0.648$). However, the relationship between suprailliac skinfold thickness and MRI was similar in both ethnic groups ($r = 0.647$, $P = 0.020$ and 0.610, $P = 0.041$). There is only a moderate correlation ($r < 0.65$) between Bodystat (BIA), tricep skinfold thickness and bicep skinfold thickness with MRI percent body fat in the Caucasian group (table 3), therefore, these variables were excluded from the multiple regression analysis. This was also the case in the Asian Indian group for Tanita (BIA) and suprailliac skinfold thickness.

Table 3. Correlation coefficients (r) of five 2C model methods of estimating percent body fat and four skinfold sites (bicep, tricep, subscapular and suprailliac) compared to the criterion measure (MRI).

	Correlation coefficients (r)			
	Caucasian (n = 11)		Asian Indian (n = 9)	
MRI vs Skinfolds	0.721	*P* = 0.006	0.821	*P* = 0.007
MRI vs Bodystat (BIA)	0.508	*P* = 0.055	0.666	*P* = 0.050
MRI vs Tanita (BIA)	0.767	*P* = 0.003	0.529	*P* = 0.111 (n=7)
MRI vs ADP	0.878	*P* ≤ 0.0001	0.796	*P* = 0.010
MRI vs HW	0.808	*P* = 0.001	0.748	*P* = 0.020
MRI vs Bicep SKF	0.584	*P* = 0.030	0.807	*P* = 0.009
MRI vs Tricep SKF	0.556	*P* = 0.038	0.852	*P* = 0.004
MRI vs Subscapular SKF	0.626	*P* = 0.020	0.810	*P* = 0.008
MRI vs Suprailliac SKF	0.647	*P* = 0.016	0.610	*P* = 0.041

Regression analysis

Regression analyses (table 4) produced one model, in the Caucasian group, that includes ADP percent body fat to predict MRI percent body fat. Stepwise multiple regression only includes the variables that make the greatest statistical contribution to the prediction model. ADP is included in the model due to its strong positive correlation with MRI (r = 0.878). The coefficient of determination (r^2) reveals that 77.1% of the variance in MRI percent body fat is associated with changes in the variable ADP. The estimated coefficient of determination for the population (adjusted r^2) reveals that 74.6% of the variance in MRI percent body fat is associated with changes in the variable ADP in the population tested. The standard error of the estimate (SEE.) is ±3.274; this value reflects the amount of

variation of the data points around the line of best fit, representing the prediction error of the model to estimate MRI percent body fat.

Table 4. Regression analyses to predict MRI percent body fat in Caucasians and Asian Indians.

Model	Intercept (A)	Slope (B)	r	r^2	Adjusted r^2	s.e.e.	*P*
Caucasian							
1. (ADP)	0.757	1.76	0.878	0.771	0.746	3.274	0.001
Asian Indian							
1. (Tricep SKF)	17.327	0.926	0.852	0.726	0.687	3.110	0.004
2. (Tricep SKF)	0.757	0.716	0.962	0.926	0.901	1.750	0.001
(HW)		0.476					

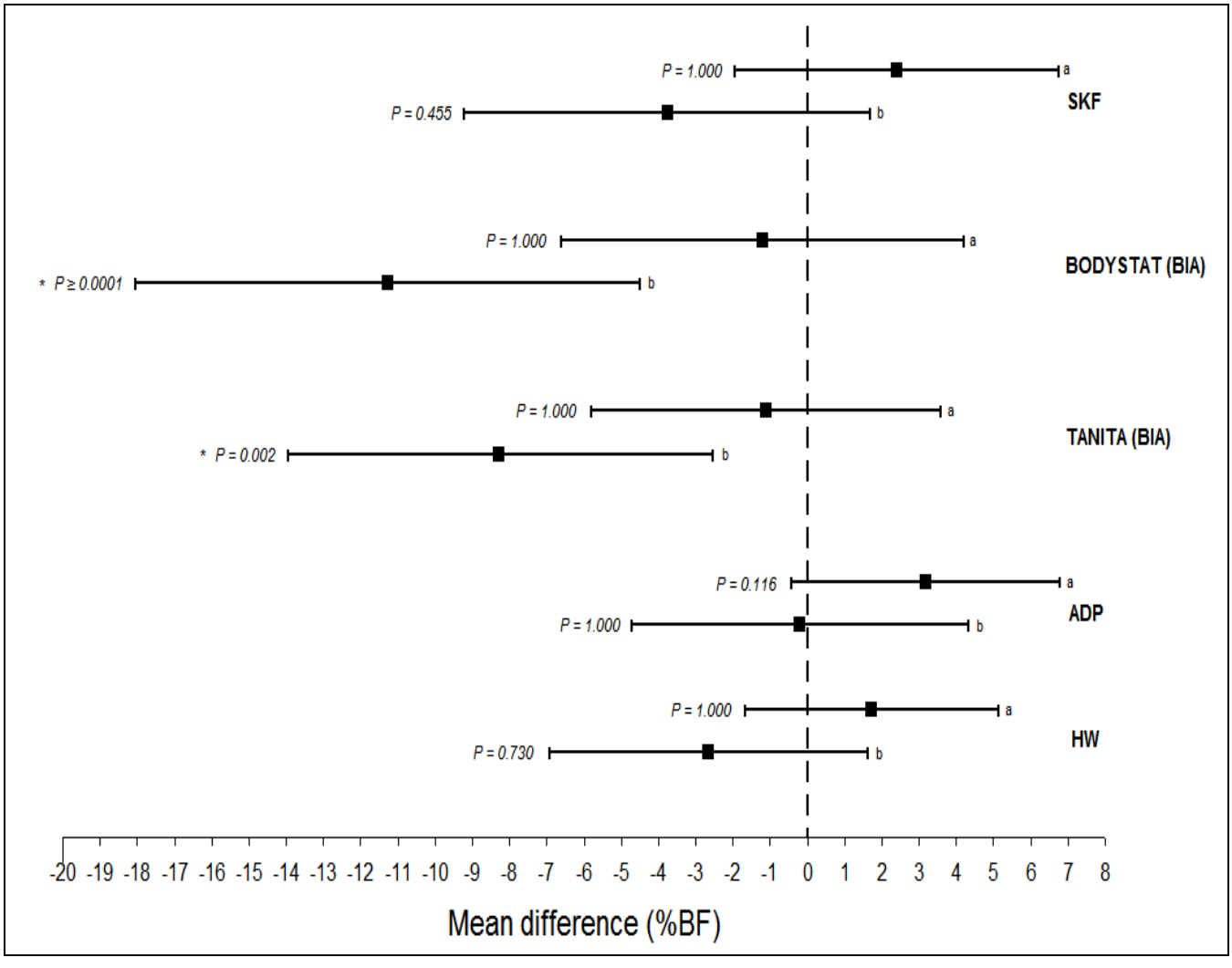
In the Asian Indian group, two models were produced: the first model includes the predictor variable tricep skinfold thickness, revealing that 68.7% (adjusted $r^2 = 0.687$) of the variance in MRI percent body fat is associated with changes in tricep skinfold thickness in the population tested. The standard error of the estimate (SEE. = 3.11) is similar to model 1 of the Caucasian group. The adjusted r^2 is greatly improved by the addition of the predictor variable HW, that demonstrates 90.1% of the variance in MRI is associated with changes in the variables tricep skinfold thickness plus HW in the population tested. The standard error of the estimate is also improved (SEE. = 1.75). Model 2 for Asian Indians was the best for predicting MRI percent body fat out of the un-presented models tested by the regression analyses.

Mean differences and confidence intervals

Mean differences and 95% confidence intervals (CI) between MRI and the five predictor estimates of percent body fat are displayed in figure 2. The mean MRI percent body fat value was subtracted from the mean percent body fat value of each 2C device to produce a mean difference for both ethnic groups, the dashed zero line indicates MRI measured percent body fat. Within groups comparison of Caucasians revealed no significant mean difference between the criterion measure and the five other devices. ADP overestimated MRI percent body fat by the greatest degree (3.19%), whilst both BIA devices, Bodystat and Tanita provided the closest measurement to MRI in the Caucasian group, underestimating percent body fat by 1.20% and 1.10%, respectively. HW was ranked as the third closest measure to MRI, and skinfolds was fourth closest. The error bars representing the 95% confidence interval show that ADP in the Caucasian group is close to being significantly different from MRI, because the error bars only just cross zero.

In contrast to the Caucasian group, BIA (Bodystat and Tanita) significantly underestimated percent body fat by 11.27 (95% CI = -4.49 to -18.05% BF; $P \leq 0.0001$) and 8.29% (95% CI = -2.44 to -14.14% BF; $P = 0.002$), respectively in the Asian Indian group. Again, unlike in the Caucasian group, ADP exhibited the least difference to MRI percent body fat with a 0.20% underestimation (95% CI = -4.72 to 4.32% BF; $P = 1.000$). HW was the second closest measure to MRI and skinfolds were ranked third.

In the Caucasian group, all estimates of percent body fat, other than BIA (Bodystat and Tanita), overestimate percent body fat compared to MRI, In contrast, percent body fat was underestimated by all 2C methods in the Asian Indian group. The large confidence intervals around the mean differences of all 2C devices against MRI show the large range of variability in both ethnic groups of which 95% of the larger parent population would fall.



(a = Caucasian, b = Asian Indian, vertical dashed line represents mean MRI value)

Figure 1. Mean differences and 95% confidence intervals for 2C devices (SKF, BODYSTAT (BIA), TANITA (BIA), ADP, HW and MRI) minus MRI.

Agreement

To further assess the validity of the models produced by the multiple regression analyses, Bland-Altman plots (Bland & Altman, 1986) were employed. Residual body composition scores of the included devices were analysed against those of MRI to determine the 95% limits of individual agreement.

Figure 2-a, demonstrates a mean difference (MRI-ADP %BF) of -3.19% (± 3.60 SD), the majority of the differences would be expected to lie ± 2 SD. Therefore, ADP produces percent body fat values 10.39% below and 4.02% above that of MRI measured percent body fat, representing a large degree of disagreement between the two methods (Bland & Altman, 1986). The solid regression line is indicative of a systematic bias, leaner individuals have estimates greater than the mean and more adipose individuals have estimates below the mean difference.

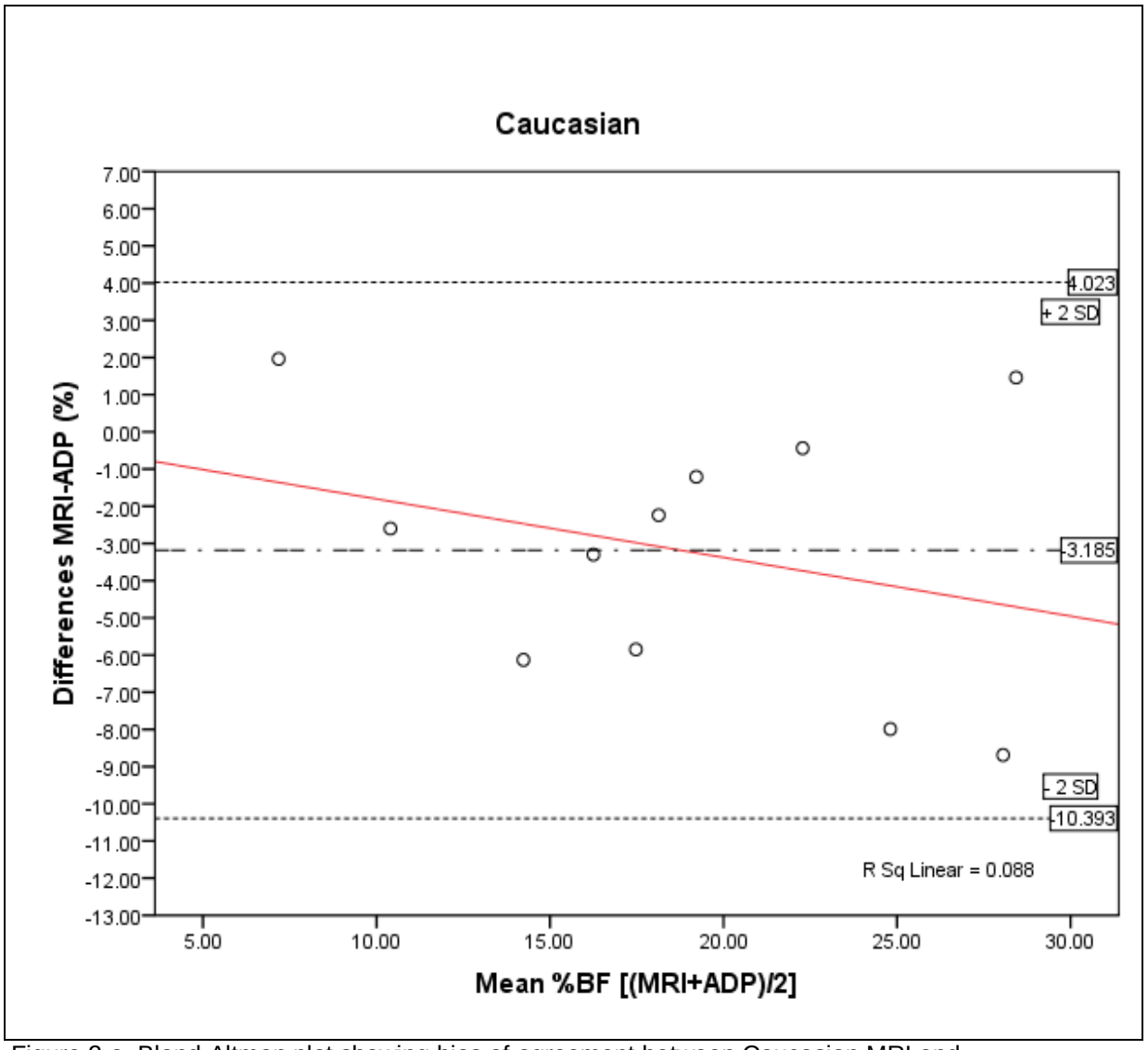


Figure 2-a. Bland-Altman plot showing bias of agreement between Caucasian MRI and ADP percent body fat values (difference between MRI and HW against their mean). Central line is group mean difference and the outer lines represent ± 2 standard deviations.

Figure 2-b, demonstrates a mean difference (MRI-HW %BF) of 4.05% (± 3.99 SD); the majority of the differences would be expected to lie ± 2 SD. Therefore, HW produces percent body fat values 3.93% below and 12.04% above that of MRI measured percent body fat. This represents a large degree of disagreement between the two methods (Bland & Altman, 1986). The solid regression line indicates there is no systematic bias. However, there appears to be an outlying data point, outside of the two standard deviations from the

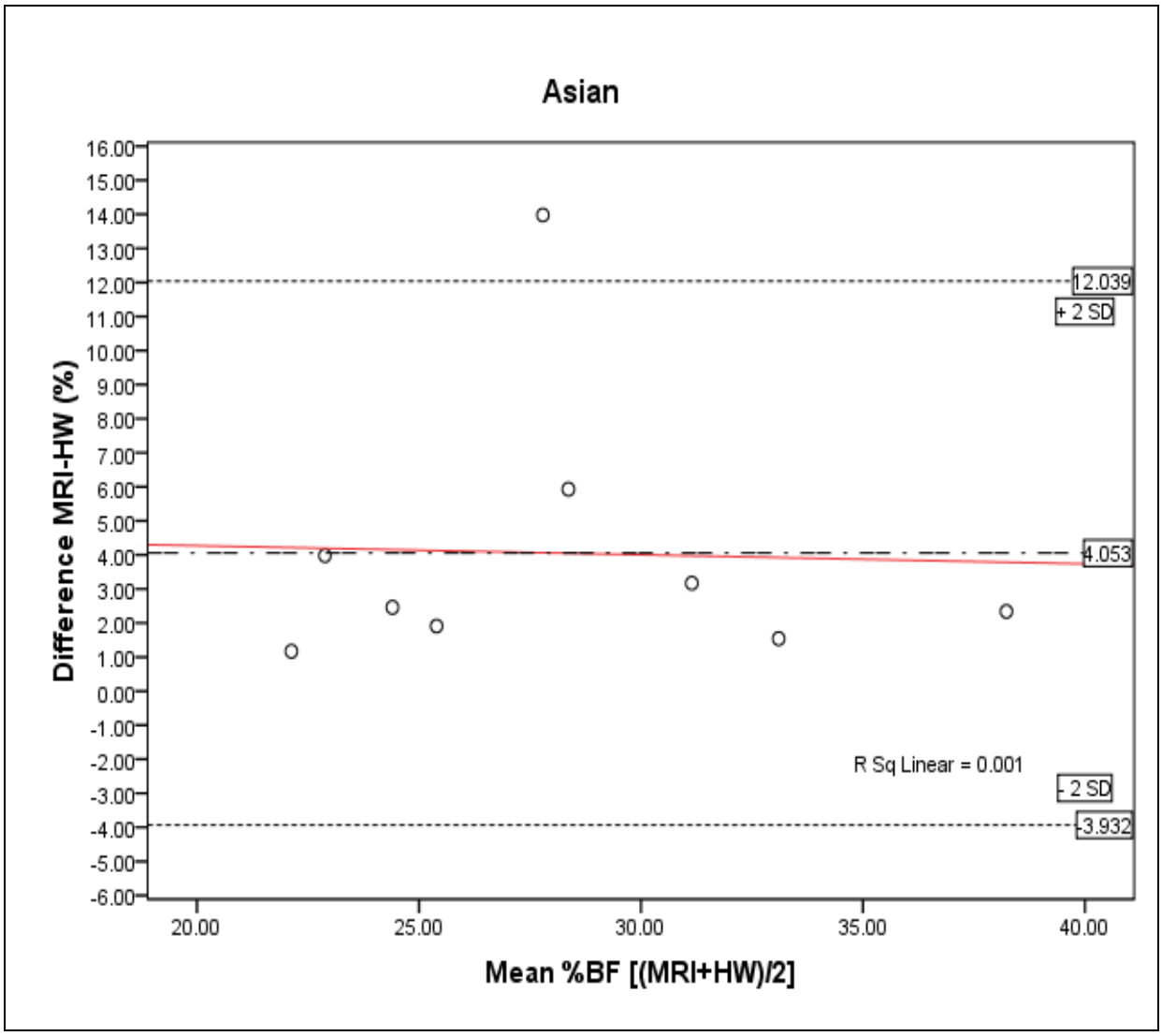


Figure 2-b. Bland and Altman plot showing bias of agreement between MRI and HW percent body fat values (difference between MRI and HW against their mean). Central line is group mean difference and the outer lines represent ± 2 standard deviations.

mean, it is likely that without this outlying value the bias and limits of agreement would be reduced.Bland-Altman plots revealed insufficient agreement between MRI and all other 2C methods.

Reliability

Table 5.A low degree of error between three repeated trials of body composition measured by all 2C devices can be observed, all trial comparisons exhibit typical error less than 0.71% BF. This suggests good

reliability of the 2C devices tested (Hopkins, 2000). Intra-class correlation coefficients show strong positive correlations between all pairs of trials. This further supports high levels of reliability of the devices tested. The final pair of trials do not show greater reliability than the preceding comparisons, this suggests that there is no improvement in reliability as a result of a familiarisation effect on both the experimenter or participant.

Table 5. Typical error of measurement and intra-class correlation coefficients between three pairs of trials for each 2C device

		1 vs 2	2 vs 3	3 vs 1
Tanita (BIA)	**Typical error % body fat**	0.34	0.23	0.37
	ICC	0.992	0.996	0.982
Bodystat (BIA)	**Typical error % body fat**	0.26	0.14	0.22
	ICC	0.995	0.998	0.993
HW	**Typical error % body fat**	0.67	0.37	0.70
	ICC	0.988	0.996	0.972
ADP	**Typical error % body fat**	0.43	0.50	0.45
	ICC	0.998	0.996	0.998

Table 6.displays typical mean error of percent body fat values, and log transformed typical error as a coefficient of variation Coefficient of variation (CV) is the ratio of the standard deviation to the mean for each pair of trials averaged to produce a mean coefficient of variation for each device. Both BIA devices have the lowest degree of typical error showing the most reliability, with BIA- bodystat ranked first for reliability. ADP is ranked third whilst HW is the least reliable. When this is expressed as a log transformed coefficient of variation BIA (bodystat and tanita) demonstrate the least variation 2.2 and 2.8%, respectively whilst ADP and

HW display 4.0% variation. All devices are sufficiently reliable as they demonstrate less than 5% variation (Hopkins 2000).

Table 6. Mean typical error of measurement (TEM) and mean TEM as a coefficient of variation for each 2C device (95% limits of agreement)

	Mean TEM (% BF) (95% CL)	**Mean TEM as CV (%) (95% CL)**
BIA-Tanita	0.32% BF (0.26-0.42%)	2.8% (2.2-3.6%)
BIA-Bodystat	0.21% BF (0.17-0.28%)	2.2% (1.8-2.9%)
ADP	0.46 % BF (0.37-0.60%)	4.0% (3.2-5.3%)
HW	0.60% BF (0.48-079%)	4.0% (3.2-5.2%)

Table 7.exhibits mean values and standard deviations of variables associated with central obesity. These are displayed due to their relationship to morbidity and mortality rates and their increased prevalence in Asian Indians (Chandalia et al, 1991 & Raji et al, 2001). They were included in order to enhance our understanding of which storage compartment (visceral or subcutaneous abdominal fat) is associated with health problems.

WHR was significantly higher in Asian Indians vs Caucasians (0.89 ± 0.53 vs 0.84 ± 0.03, P = 0.016). Hip circumference was lower in the Asian Indian group but not significantly (97.67± 5.66cm vs 100.48 ± 7.18cm, P = 0.335). Waist circumference was higher in the Asian Indian group but the difference was insignificant (87.58 ± 9.72cm vs 84.54 ± 7.38cm, P = 0.426). Subcutaneous abdominal fat as a percentage of body mass and

when represented as a percentage of adiposity was significantly higher in the Asian Indian group, ($P \leq 0.0001$ and $P = 0.029$, respectively). When visceral fat was presented as a percentage of body mass it was significantly higher in the Asian Indian group ($P \leq 0.0001$), but when presented as a percentage of adiposity the mean difference was non-significant ($P = 0.064$).

Table 7. Means and standard deviations of variables associated with central obesity: waist to hip ratio (WHR), hip circumference, waist circumference, subcutaneous abdominal fat as percentage of body mass, subcutaneous abdominal fat as a percentage of body fat and visceral fat as a percentage of body fat.

	Caucasian (n=11)	**Asian (n=10)**	**Sig**
WHR	0.84 ± 0.03	0.89 ± 0.53*	P = 0.016
Hip circumference (cm)	100.48 ± 7.18cm	97.67 ± 5.66	P = 0.335
waist circumference (cm)	84.54 ± 7.38cm	87.58 ± 9.72	P = 0.426
Subcutaneous abdominal fat % body mass (%)	3.38 ± 1.73	6.81 ± 1.87*	P ≤ 0.0001
Subcutaneous abdominal fat % adiposity (%)	18.83 ± 4.05%	22.57 ± 3.08*	P = 0.029
Visceral fat % body mass (%)	1.74 ± 1.15	3.66 ± 0.92*	P ≤ 0.0001
Visceral fat % adiposity (%)	9.37 ± 3.94%	12.33 ± 2.75	P = 0.064

5. DISCUSSION

Overview

The objective of the current investigation was to test the validity of simple methods of body composition when used with Asian Indian males. This was done by assessing the validity of these simple methods against a criterion measure (MRI) in both Asian Indian and Caucasian males. Unfortunately, there is little data on the validity of body composition techniques in Asian Indians making a comparison of the current Asian Indian data difficult. There is also limited data validating 2C methods against MRI; in the majority of studies the most advanced criterion measure is DXA. The following will discuss the findings of the current study in relation to previous literature and their implications.

Major Findings

The two-compartment model consistently underestimated body composition compared to MRI in the Asian Indian group, whilst the majority of 2C methods overestimated body composition of the Caucasian group. The majority of these differences, however, were not significantly different from MRI,apart from BIA (Tanita and Bodystat) in the Asian Indian group ($P = 0.002$ and $P = 0.0001$, respectively). There were significant positive correlations between MRI and skinfolds, MRI and ADP and MRI and HW in both ethnic groups. BIA (Bodystat) in the Caucasian group and BIA (Tanita) in the Asian Indian group were not significantly correlated. Regression analysis revealed that MRI measured body composition can be predicted for Asian Indians by tricep skinfold thickess and HW with a

low prediction error (adjusted $r^2 = 0.90$; SEE = 1.75). Bland-Altman plots, however, reveal there is an unacceptably wide range of individual variability between the 2C model variables included in the regression model (ADP and HW) and MRI percent body fat estimates.

According to these findings, the first null hypothesis: there will be no significant difference between percent body fat values estimated by the 2C methods (ADP, HW and SKF), is accepted, with the exception of BIA for the Asian Indian group, which is rejected. The first alternate hypothesis: there will be a significant difference between percent body fat values estimated by the 2C methods (ADP, HW and SKF), is rejected, with the exception of BIA for the Asian Indian group, which is accepted.

The second null hypothesis, that there will be no significant linear relationship between percent body fat scores estimated by the 2C model methods compared to the criterion MRI, can be accepted for BIA (Bodystat) in the Caucasian group and BIA (Tanita) in the Asian Indian group. The second null hypothesis, however, is rejected for SKF, ADP and HW in both ethnic groups. The second alternate hypothesis: There will be a significant linear relationship between percent body fat scores estimated by the 2C model methods compared to the criterion MRI, is rejected for the Bodystat BIA device in the Caucasian group and the Tanita BIA device in the Asian Indian group. The Second null hypothesis is accepted for the SKF, ADO and HW in both ethnic groups.

The third null hypothesis: There will not be sufficient agreement to infer validity between percent body fat scores estimated by the 2C model methods (ADP, BIA, HW and SKF) can be accepted within both ethnic groups. The third Alternate hypothesis: There will be sufficient agreement to infer validity between percent body fat scores estimated by the 2C model methods (ADP, BIA, HW and SKF) is rejected in both ethnic groups.

Findings related to literature

Physical characteristics of the participants (Table 1) revealed that Caucasians were older with a greater age range than Asian Indians. Caucasians were slightly taller and considerably heavier than Asian Indians. As a result of this increased body mass of Caucasians, BMI was greater in the Caucasian group (26.19 ± 3.34kg/m² vs 25.38 ± 3.85 kg/m²). BMI classified both groups in the overweight-pre obese category, according to WHO (2006). Despite this, MRI measured percent body fat was over 12% higher in the Asian Indian group (17.18 ± 6.49% vs 29.91 ± 5.31%). This places these Asian Indians in the obese category and the Caucasians in this study within the mid range of recommended body fat levels (Lohman et al, 1997). Thus, BMI, when compared to MRI body composition measures, is misclassifying both groups. According to BMI, Caucasians are at greater risk and Asian Indians are at less risk of obesity related disorders, but percent body fat as measured using MRI suggests the reverse. Similar observations were made by Banerji et al (1999) in a comparison between BMI and body composition aquired via

CT imaging. This highlights the benefit of body composition measures over BMI.

Within groups comparison of the mean differences of Caucasian percent body fat (Figure 1) reveals that skinfolds, ADP and HW all overestimate percent body fat compared to MRI (2.40%, 3.19% and 1.73%, respectively), however, both BIA devices (Bodystat and Tanita) underestimate percent body fat (-1.20% and 1.10%) and showed the least error compared to MRI. None of these differences were significant in the Caucasian group and all 2C devices, other than Bodystat (BIA) ($r = 0.508$, $P = 0.055$), correlated quite strongly and significantly with MRI (table 3.). ADP and HW exhibit the strongest correlation with MRI ($r = 0.878$, $P \leq 0.0001$ and $r = 0.808$, $P = 0.001$, respectively). These findings suggest that there is a non-significant degree of error and a similar relationship between the 2C devices (skinfolds, Tanita [BIA], ADP and HW) and the criterion measure MRI in the Caucasian group. This suggests that they are valid estimates of body composition, however the degree of error is still larger than to those observed in past research. Wagner et al (2000) found a mean difference of 0.25% body fat from HW compared to DXA.

The strong correlation between the 2C devices and MRI renders them suitable for inclusion in multiple regression analyses to derive a model for the prediction of MRI percent body fat. Similar findings have been reported by Radley et al (2003), who found that the 2C method of ADP was found to be non-significantly overestimated (0.67%) and highly correlated (r =

0.84) in a comparison with body fat measures by the reference DXA method. Duz et al (2009) reported that BIA underestimated body fat and the closest to the criterion measure (DXA), skinfolds had a greater difference (4.8 ± 0.7% and 6.1 ± 0.5%, respectively), which is similar to the current study. However, at odds with the current study, skinfolds underestimated the true adiposity and both devices were significantly different from the criterion. These contrasting findings could be due to the measurement techniques, BIA was acquired by a hand held device to estimate percent body fat which only measures body water of the upper body, in the arms and across the chest (Deurenberg & Deurenberg-Yap, 2002) and is less valid than hand-to-foot BIA used in the current study, which is likely to give a better understanding of total body impedance. Total body fat from skinfolds was obtained by the three site equation of Jackson and Pollock (1978), that includes a skinfold site of the lower body, where as the current study used the four site equation of Durnin and Womersley (1974), which only uses upper body skinfold sites. Furthermore, the smaller sample used in the current study is likely to decrease the chance of significant differences being observed. The large confidence intervals exhibited in both ethnic groups (Figure 2), show that 95% of the population tested fall within a large range around the mean difference, suggesting there may be issues with the sample size. Large confidence intervals can be representative of a small sample size, a greater number of participants would likely reduce the confidence interval. Therefore, a small sample size, like that of the current study increases the

chances of producing a non-significant difference between the predictor and criterion variables.

This may be demonstrated as follows: Looking at Figure 2 it is noted that the Asian MRI-ADP has the closest relationship to being 'the same' measured values. Applying Cohen's (1989) power prediction equation $n = (2(SD)^2*(Z_\alpha+Z_\beta))/\Delta^2$ (where alpha = 0.05 and beta = 0.80, SD = SD_{ADP}-SD_{MRI}, and delta = a minimal detectable change of 0.01 %BF as measured by both ADP and MRI) we find a value of n = 10.2; this needs to be rounded to 11 to represent 'whole' participants, divided by two = 5.5 per group, rounded to 6 per group As such, even with Tanita having less participants (n = 7), it is still greater than 6 therefore, if there was a significant difference to be found it is likely have already been shown.

Looking at Figure 2 it is also noted that the Caucasian MRI-ADP has the closest relationship to being significantly different while remaining non-significant. Applying Cohen's (1989) power prediction equation $n = (2(SD)^2*(Z_\alpha+Z_\beta))/\Delta^2$ (where alpha = 0.05 and beta = 0.80, SD = SD_{ADP}-SD_{MRI}, and delta = a minimal detectable change of 0.01 %BF as measured by both ADP and MRI) we find a value of n = 171.2; this needs to be rounded to 172 to represent 'whole' participants, divided by 2 = 86 per group. As such, the strong trend indicated by Caucasian MRI-ADP Mean Diff $\pm$ 95% CI (Figure 2) indicates that according to Cohen (1989) a significant difference should be found when n = (2 groups of) 86. This

indicates that a further 76 Asian Indian and 75 Caucasian subjects would be needed to confirm either significant or non-significant findings within the context of this project (see Appendix G for calculations).

In contrast to the Caucasian group, within groups comparison of the Asian Indian group (Figure 2) reveals that all 2C devices underestimate percent body fat compared to MRI. BIA (Bodystat and Tanita) were the only devices to produce significant differences compared to MRI (-11.27%, $P \leq 0.0001$ and -8.29, $P = 0.002$). Skinfolds, ADP and HW exhibited non-significant differences (-3.76%, -0.20% and 2.65%) compared to MRI. Correlation analyses (table 3.) reveals that skinfolds, ADP and HW all display significant positive relationships to MRI ($r = 0.821$, $P=0.007$; $r = 0.796$, $P = 0.010$; and $r = 0.748$, $P = 0.020$, respectively). Bodystat (BIA), although significant, had a weaker correlation with MRI ($r = 0.666$, $P = 0.050$). Tanita (BIA) did not correlate significantly with MRI ($r = 0.529$, $P = 0.111$). The non-significant degree of error between skinfolds, Tanita (BIA), ADP and HW compared to MRI suggests that these 2C devices are accurate estimates of body composition, however, the quantitative differences are still quite large.

The significant positive correlation between skinfolds, Bodystat (BIA), ADP and HW means they could contribute to a prediction model of MRI body composition derived through regression analysis. The significant difference between mean scores of percent body fat and the non-significant relationship between Tanita (BIA) and MRI in the Asian Indian

group, suggests it may not be a valid device for body composition estimation compared to MRI in this population. However, the small sample size (n=7) makes it difficult to confirm or deny such a notion, this outcome may have also been different if an equal number of participants were measured with this device compared to the remaining 2C devices. The ADP confidence interval error bars only just cross zero; a significant difference is displayed if zero does not fall between the error bars. If the sample size was greater the confidence intervals may not cross zero and thus a significant difference may have been observed between ADP and MRI in the Caucasian group. This is highlighted by the findings of Bhat et al (2008) who reported smaller mean differences (skinfolds,1.6% and BIA, 1.4%) than the current study compared to a criterion (deuterated water) method. The current study found non-significant differences in values greater than those expressed by Bhat et al (2009), this is like to be due to the greater sample size in their study (n = 145 compared to n = 21 in the current study). Bhat et al (2009) may have found a greater difference between skinfolds and the reference measure and between BIA compared to the reference measure if they used a more advanced criterion such as MRI, which was used in the current investigation.

Table 3 also exhibits correlations of the four skinfold sites (bicep SKF, tricep SKF, subscapular SKF and suprailliac SKF) that contribute to the sum of four skinfolds (Durnin & Womersley, 1974) to produce the skinfolds estimate of body composition. In the Caucasian group, all four skinfold sites significantly and positively correlate with MRI percent body fat,

however, all correlations are moderate, none reaching greater than r = 0.647. In the Asian Indian group, Suprailliac SKF correlates least with MRI (r = 0.610), but remains significant. All other skinfold sites have strong, significant positive relationships with MRI, all above r = 0.807. These strong correlations in the Asian Indian group warrant their inclusion in multiple regression analysis as they may contribute to the prediction equation. Past research has identified individual skinfold site as useful variables in regression analysis (Warner et al, 2004; Volz & Ostrove, 1984).

In the Caucasian group, regression analysis (Table 4) produced one model, determining that only ADP was sufficient for predicting MRI percent body fat. In this case no other variables added to the prediction model in the Caucasian group. The regression equation ($Y= a_y + [b_y][x]$) to predict MRI percent body fat is, therefore, as follows:

Caucasian MRI% BF = 0.757 + (1.76 × ADP% BF)

The adjusted r^2 reveals that 74.6% of the variance in MRI percent body fat is associated with changes in ADP, showing moderate to strong correlation between the prediction model and MRI measured body composition. The standard error of the estimate (SEE.) is 3.274% body fat which equates an error range of 6.548%, Lohman (1992) developed standards to evaluate the prediction error (SEE.) of equations to predict body composition. According to Lohman (1992) a SEE. of 3.274% is rated as good to very good. The significance value ($P \leq 0.0001$) in Table 4

shows that the model is significantly better at predicting MRI percent body fat than a ‘best guess’ made without the model, suggesting the model has predictive value in Caucasian males outside the population tested. However, it is important to note the small sample size used to create this model (n=11).

In the Asian Indian group, regression analysis produced 2 prediction models (Table 4). Model 1, includes tricep SKF. The regression equation is as follows:

Asian Indian MRI% BF = 17.327 + (0.926 × tricep SKF)

Adjusted r^2 shows that 68.7% of the variance in MRI is associated with changes in tricep SKF, which is a moderate correlation. The SEE is 3.11%, rated good to very good (Lohman, 1992), with a significance of P = 0.004. The predictability of model 1 in the Asian Indian group, is greatly improved and the standard error of the estimate is reduced (SEE = 1.75% BF), by the addition of HW in model 2. According to Lohman (1992), a SEE of 1.75, is rated as ideal. The improved regression equation is as follows:

Asian Indian MRI% BF = 0.757 + (0.716 × tricep SKF + 0.476 × HW)

Adjusted r^2 reveals that 90.1% of the variance in MRI is associated with changes in tricep SKF and HW, $P \leq 0.0001$. From the regression models,

ADP percent body fat can be used to predict MRI percent body fat in the Caucasian sample. In Asian Indians, tricep SKF measurement can predict MRI percent body fat but the inclusion of HW percent body fat produces a stronger prediction equation with an ideal prediction error (1.75% body fat). The use of individual skinfold sites in regression models have been used to predict DXA measured body composition by Warner et al, (2004) who reported body mass, abdominal skinfold and thigh skinfold predicted fat free mass (r = 0.98, SEE = 1.1kg).

It is important to assess the bias of agreement between the devices used in the prediction models and MRI, as a non-significant relationship and strong correlation from regression analyses can demonstrate valid use of the prediction to assess criterion body composition. Bland-Altman analysis provides important information that a regression analysis will not detect (Williams & Bale, 1988). Unacceptably high limits of agreement between a predictor and criterion mean the predictor is not a valid replacement for the criterion (Williams & Bale, 1988)

Bland-Altman plots (Figure 2 a and b) show bias and the reference range of the differences between individual subject values for MRI and the 2C devices included in the regression models. ADP, used to predict MRI in the Caucasian group (Figure 2-a) shows a bias of -3.18% between the differences of the mean MRI-ADP measures. Limits of agreement show a 95% chance that a participants actual MRI measured body fat percentage will fall between -10.39% and 4.02% body fat of their ADP body fat

percentage. These limits indicate an unacceptably wide range of individual variability between ADP and MRI percent body fat estimates in Caucasians. According to Brodie (1988), limits of agreement greater than 2% body fat are unacceptable. There also appears to be a systematic bias, there is greater error in individuals who are more lean and those with higher levels of body fat. HW, used to predict MRI body fat in the Asian Indian group (Figure 2-b), displays a bias of 4.05% between the differences of the mean MRI-HW measures. Limits of agreement show a 95% chance that an individuals actual MRI measured body fat will lie between -3.93% and 12.04% of their HW value. These are unacceptably high limits of variability between the two devices, (Brodie, 1988). It is important to note an outlying data point displayed in Figure 3-b; without this outlying value the mean difference and bias and agreement would be reduced. Appendix H, displays figure 2-b, with the outlying data point removed. Although the bias is reduced from a mean of 4.05% to 2.81% and the limits of agreement are reduced from (-3.93% to 12.04%) to (-0.28% to 5.90%), the variability between the two devices remains unacceptably high (Brodie, 1988).

An interesting observation is the greater mean difference and confidence intervals of the 2C devices versus MRI in the Asian Indian group. It is possible that this relates to ethnic variation of fat free mass density. Schutte et al (1984) reported that different ethnic groups have different fat free mass densities. As Caucasians were used to develop the 2C model based on the findings of Siri (1956) and Brozek et al (1963) a smaller

mean difference between the 2C methods and MRI in the Caucasian group could be expected due to the population specific nature of the 2C model, as present in the current findings. Thus, the greater mean difference in percent body fat produced by the 2C devices compared to MRI in the Asian Indian group (apart form ADP) could be due to a difference in fat free tissue density producing less valid estimates of body composition. According to Schutte et al (1984), Black individuals had a higher fat free mass density resulting in an underestimation of percentage body fat. Based on the findings of Schutte et al (1984), it is possible that the underestimated percent body fat seen in all 2C methods compared to MRI in the Asian Indian group could be due to a higher fat free mass density in the Asian Indian group. This cannot be said for BIA, as it does not assume a constant density for the fat free body. It will not be until the fat free density of Asian Indians is quantified through such techniques as DXA to measure bone density or cadaver studies to assess the density of all lean tissues, that such a notion can be supported. It is likely, however, that other factors could also be contributing to an underestimated body composition measured by the 2C devices in the Asian Indian group. These may be explained by the principles underpinning the different 2C techniques.

BIA is based on the principles of electrical conductivity, combined with basic assumptions of the geometric shape of the body and of the relationship of impedance (opposition to flow of current) to the volume and length of the conductor (Heyward & Wagner 2004). Differences in body

proportions and body fat deposition could be causing the conflicting estimates from the BIA devices between the two groups, observable in the current findings. Ethnic differences in body fat deposition and body proportions have implications for field methods such as BIA and SKF (Deurenberg & Deurenberg-yap, 2001; Wagner & Heyward, 2000). Ward et al (2000) in their review of previous body impedance studies, identified ethnic differences in body impedance, that would invalidate BIA derived body composition in different ethnic groups. The assumption that the body is a perfect cylinder, as made by the whole-body, tetrapolar BIA model, is not entirely correct. The body more closely resembles five cylinders (two arms, two legs and a trunk), excluding the head (Kushner, 1992). The resistance to the current will differ in the various body segments due to their variable length and cross-sectional area. This means that varying body proportions of different ethnic groups, seen in Asian Indians compared to Caucasians (WHO, 2009) will also impact on impedance value. When impedance is expressed in terms of body volume, the product of the equation will be inaccurate as there are different sized cylinders, contributing to the resistance of the current.

ADP had the greatest difference compared to MRI measured body fat in the Caucasian group. The mean difference between Asian Indian ADP and MRI was the smallest out of all 2C methods compared to MRI in both ethnic groups. As the 2C model was developed utilising data from Caucasian cadavers (Siri, 1956) it could be expected that the 2C devices would produce more valid data, compared to MRI, for the Caucasian

rather than Asian Indian participants. A possible reason for this could be the effect of iso-thermal air (constant temperature) surrounding the participants in the BodPod chamber. Fields et al, (2004) state that the BOD POD operates under adiabatic conditions (no loss or gain in temperature) allowing for changes in air temperature due to the presence of a subject in the chamber. Around the surface of the skin and in the subject's lungs the air is isothermal which is more compressible than adiabatic air and is thus corrected for by measuring or predicting thoracic gas volume during testing and applying a surface area artefact to the body volume equation (McCrory et al, 1995). Interestingly, McCrory et al (1995) reported that body surface area was significantly related to a reduced percent body fat. The Asian Indian participants seemed more averse to wearing tight fitting clothing than the Caucasian group, although this was a requirement of the protocol, some Asian Indian Participants did not comply. Loose fitting clothing is known to produce a greater negative volume effect due to the iso-thermal air trapped within it (Fields et al, (2004), resulting in lower body fat reading. This may partly explain why Caucasian ADP body fat was overestimated and Asian Indian ADP body fat was underestimated when compared to MRI. However, as there is little evidence of the validity of ADP in Asian Indians, it is difficult to explain why ADP was the most valid in terms of mean difference compared to MRI in the Asian Indian group. The subjective observation of reduced participant compliance in the Asian Indian group in terms of clothing is one that should be considered in future testing, where participants are required to

wear minimal or revealing attire for valid measurement, but may be less comfortable to do so due to socio-religious reasons.

Table 2 shows that the Asian Indians in this study had greater proportions of body fat than Caucasians when measured by any 2C device as well as the criterion measure MRI. Apart from Bodystat (BIA) percent body fat, all other devices produced significantly greater estimates of percent body fat in the Asian Indian group, despite the greater body mass of the Caucasian group. Therefore the Caucasian group must have been heavier than the Asian Indian group because of a greater fat free mass, rather than fat mass.

The consistently underestimated 2C model percent body fat in the Asian Indian group, suggests that the 2C devices could mis-classify Asian Indians within a normal range of body fat, when true measures of body composition (MRI) could classify them as overweight or obese, thus, potentially allowing early indicators of obesity related disorders to go unrecognized and untreated. This is a worrying notion when considering high prevalence of obesity related disorders in Asian Indians, highlighting the importance of validation of 2C model methods of body composition measurement.

A current issue surrounding obesity is the fat distribution of Asian Indians, Table 7 displays variables associated with centralised fat storage contrasted between both ethnic groups. Asian Indians had a significantly

greater WHR than Caucasians. The tendency of Asian Indians to store truncal fat is often reflected by an increased waist to hip ratio compared to other ethnic groups (McKeigue et al, 1991; Singh et al, 1995). Asian Indians had significantly greater subcutaneous abdominal fat when expressed as a percentage of body mass ($P \leq 0.0001$). It is important, however, to analyse this variable as a percentage of body fat, because a greater lean mass would increase total body mass, resulting in a reduced subcutaneous abdominal fat when expressed as a percentage of body mass. Subcutaneous body fat as a percentage of adiposity was also significantly greater in the Asian Indian group. There is confounding literature as to whether visceral or subcutaneous body fat is contributing to a greater risk of obesity related disorders in Asian Indians (Chandalia et al, 1999). The current study found visceral fat as a percentage of body mass was significantly greater in the Asian Indian group ($P \leq 0.0001$). But when visceral fat was presented as a percentage of total adiposity the difference was not significant ($P = 0.064$). This suggests that Asian Indians stored a greater proportion of abdominal fat subcutaneously, in relation to total fat, compared to Caucasians. This suggests that the greater prevalence of obesity related disorders in Asian Indians could be due to subcutaneous not visceral, abdominal body fat. These data can be applied to the findings of Valsamakis et al (2004) who suggested that subcutaneous abdominal body fat (measured by MRI) plays a greater role in the development of obesity related type II diabetes. At odds with these findings, Caprio et al (1993), reported significant positive correlations of visceral fat and metabolic risk factors. The current study did not ascertain cardiovascular

and metabolic risk factors and thus can only suggest a link between increased subcutaneous abdominal fat storage and health risk in Asian Indians in the current study based on previous research.

Limitations

There are a number of limitations surrounding the sample used in the current study. Firstly, the sample was small, It is important to use a sufficient sample to gain statistical power, which is the probability that a study will find a significant statistical effect (Atkinson, 2005). There can be difficulties of acquiring a statistical significance as the related low degrees of freedom make it hard for a test statistic to be larger than the critical value (Atkinson 2005). Sample size is also an issue when using regression analysis, generally, large samples (n = 100- 400) are needed to maintain that the data represent the population for whom is being tested (Heyward & Wagner, 2004), and in this instance was not achieved, Caucasians (n = 11) and Asian Indians (n = 10), thus making the current findings difficult to generalise to the greater public. Moreover, In the Asian Indian group, not all ten participants were measured using all the 2C methods, therefore, the effects a small sample size may have been exaggerated as a result. Secondly, the sample was of males and can only be generalised to that gender group. Thirdly, acceptance criteria included that participants were sedentary, however, no data was gathered in order to assess the participants level of physical activity. This could result in participation from those who are physically active and those who are sedentary, potentially causing extremes between participants, making

generalisability to a particular population difficult. Another limitation is the pre-testing protocols instructed to the participants before testing took place. They were asked to be fasted and refrain from exercise 4 hours prior to testing, however, the only follow up was to ask the participants if they had conformed to the pre-testing protocol. Failure to conform to such guidelines may have distorted results. Other outside factors that may alter findings is the hydration level of participants. This was not accounted for in the current study and therefore the effects of hydration on the outcome of body composition values are unknown. Hydration is a very important factor when assessing body composition using the 2C model, as a water content of 73% is assumed to be constant within the body (Heywood & Wagner, 2004), particularly when using BIA, as it measures total body water to estimate body composition.

A further limitation is that the current investigation predicted thoracic gas volume (TVG) of the lungs, required for the calculation of ADP measured body composition, based on age and height tables instead of using thoracic gas measurements. The prediction tables do not account for any ethnic differences in TGV, and are less accurate than taking measurements via the BodPod software. A similar problem arises from the prediction of residual lung volume, required to calculate body composition by HW. The current study predicted residual volume from FVC values obtained via spirometry. The use of spirometry to predict residual lung volume could lead to an underestimated residual volume if the FVC measurement is inaccurate, potentially resulting in invalid body

composition estimates. Further to this, body hair can alter the accuracy of the data when estimating body composition from ADP and HW. Future prediction equations obtained on a greater sample size should be cross validated to determine the predictive accuracy of their application in practice

Implications

The results of this investigation indicate the potential for simple 2C model methods to predict an advanced reference measure of body composition (MRI) in Asian Indian and Caucasian males. This suggests ADP for Caucasians and Tricep skinfold thickness and HW for Asian Indians, are useful tools for the estimation of MRI percent body fat. This is of particular importance as the 2C model has not been validated using so many devices, nor have they been validated by such an advanced reference measure as MRI in the Asian Indian population. Thus, these less expensive alternatives could be used to accurately estimate body composition and in turn health risk in this population. However, HW and ADP are not necessarily accessible devices. They may be cheaper alternatives to a number of more advanced reference methods but access is mostly limited to hospitals and institutions of Higher education. Skinfold callipers are the cheapest and most accessible 2C device in the investigation, however the use of one skinfold measurement to predict total body fat measured by MRI may not be a valid alternative. Although regression analysis produced a prediction equation using tricep skinfold

thickness, it is unclear whether changes in tricep skinfold thikness would replicate total body fat.

Future research

In light of the limitations, a number of future recommendations can be made from the current study. Future body composition validation research into the Asian Indian population, must use a sample size representative of the population under investigation. Females should be included as participants in future research into a related topic by the current investigation, this will provide data on the Asian Indian population as a whole, increasing the generalisability of the findings. Moreover, the use of physical activity questionnaires, or more advanced techniques such as accelerometry should be employed to quantify physical activity level as this information is of great value to body composition research. Future research may benefit from measuring rather than predicting TGV for ADP estimates of body composition, this can also be said for measuring RLV when estimating body composition by HW. Measuring TGV would account for any ethnic differences and added error as a result of predicting this value, increasing the validity of the body composition values produced by the BodPod. To avoid invalid RLV estimates, closed circuit helium dilution can be used to measure RLV. These measurements, however, are more time consuming and expensive The simplicity of predicting these values may outweigh the benefits of measuring, considering they were related enough to predict MRI body composition in the current study. Predicted versus measured RLV and TGV may make for an interesting comparison

for future 2C model validation studies of Asian Indians. A further area for future investigation would be participant hydration levels. Due to the assumptions of the hydration of the fat free body (73% water) made by the 2C model, it would be of great benefit to quantify hydration. This could be accomplished through the measurement of urine specific gravity.

The importance of body composition validation in the Asian Indian population is clear due to the increased prevalence of obesity and subsequent disorders. Cross validation of prediction equations is required to assess the predictive accuracy to justify their use on the greater Asian Indian population.

Conclusion

To conclude, multiple regression analysis determined the potential use of tricep skinfold thickness and HW to predict percent body fat as measured by MRI, with a low prediction error in Asian Indians. According to the data, in Caucasians, the best 2C device for predicting MRI body composition is the BodPod (ADP) device. Regression analysis, however revealed this prediction had less predictive accuracy than that of the prediction model produced for the Asian Indian participants (adjusted $r^2 = 0.746$ and 0.901, respectively). Despite strong correlations and non-significant differences displayed between the mean values of the 2C model devices included in the prediction equations and MRI percent body fat, bias and limits of agreement were unacceptably high. This may not have been the case if a

larger, more generalisable sample were investigated. The need for further body composition validation in the Asian Indian population is clear due to the increased prevalence of obesity and subsequent disorders, and the distinct lack of validation data currently published.

6. APPENDICES

Appendix A: Testing protocol of body composition measurement

MRI

MRI measured body fat, skinfold analysis, BMI measurement, waist circumference and hip circumference were obtained at the Hammersmith hospital by John McCarthy as part of an existing MRI research project (ref. McCarthy, REC Ref. 06/Q0411/173). Participants, once fully aware of the aims, procedures and outcomes of the study, signed informed consent. Prior to testing they were required to fill in a metal check form so that the researcher and MRI staff were fully aware of any potential health hazards or items that may degrade image quality. Any metal in the body, such as a piece of jewellery or an implant, could cause serious harm whilst being scanned.

Participants were given ear defenders in accordance with standard health and safety procedures when using magnetic resonance sacanners, it was also made clear that they can communicate with the technicians during testing via intercom. Participants were given an alarm buzzer to sound at any time if they become worried or uncomfortable during the measurement, as at any time testing can be stopped. IMCL and IHCL were measured using MR spectroscopy (MRS) and whole body adiposity was measured using MRI.

Firstly the liver was scanned (using MRS) ; participants were required to lie still in a supine position on the motorised scanner bed (can move freely through the magnet). The participant was moved into the magnet for the scan (lasting approximately 10 minutes). Then the bed was moved from out of the magnet, the participant remains still whilst the MR staff (radiographers and research team) place a focal coil around the subject's left calf muscle (lasting approximately 10 minutes), foam pads are placed under the legs for comfort. Finally, the participant was required to lye on their front whilst the whole body is scanned. The entire body was moved through the magnet on the scanner bed (lasting approximately 20 minutes). In total the scan lasts approximately 40 minutes; this includes changes of position.

Skinfold analysis

Participants are required to wear shorts and remove any clothing on their top half that may make measurements at any of the sites difficult. Measurements were taken on the right side of the body using a Harpenden skinfold caliper. Measurements were taken according to Durnin & Womersley (1974) at four sites: bicep, tricep, subscapular and suprailliac. Each site was carefully identified and marked with a water soluble pen. Each site was measured consecutively in the same order until three skinfold measures had been acquired at each site. The average of the three measures was taken and used as the corresponding skinfold thickness for that site.

Tanita (BIA)

Participant information is manually entered into the device via the main control panel, including; gender, height, age and body build. Wearing minimal clothing the participant is required to step onto the platform at the base of the device, carefully placing their feet on the electrode plates. The participant is required to stand still whilst a measurement of weight is acquired. The participant then grasps the handles, one in each hand, ensuring a tight grip around the electrodes of the handles. Whilst stood still, with arms down by their side slightly abducted from the body, a current is passed through the hands and feet around the body, for approximately 5 seconds. After a beep is sounded the participant can replace the handles on their mounts and step off the platform. Data is displayed on a printout, from the devices internal printer.

Bodystat (BIA)

The participant must be in a supine position for at least 5 minutes prior to testing. The current investigation allowed 10 minutes in a supine position to ensure body fluids had settled. During this time the participant can be prepared for measurement. The device has 4 electrodes shared between 2 wires running from the device. Sensor (proximal) electrodes are placed on the dorsal surface of the wrist, the upper border of electrode bisects the styloid process of the ulna and radius, and the dorsal surface of the ankle, the upper border of electrode bisects the medial and lateral malleoli. Source (distal) electrodes are placed at the base of the second or third

metacarpal-phalangeal joints of the hand and foot. Ensure at least 5 centimeters (cm) between proximal and distal electrodes. see figure 3.

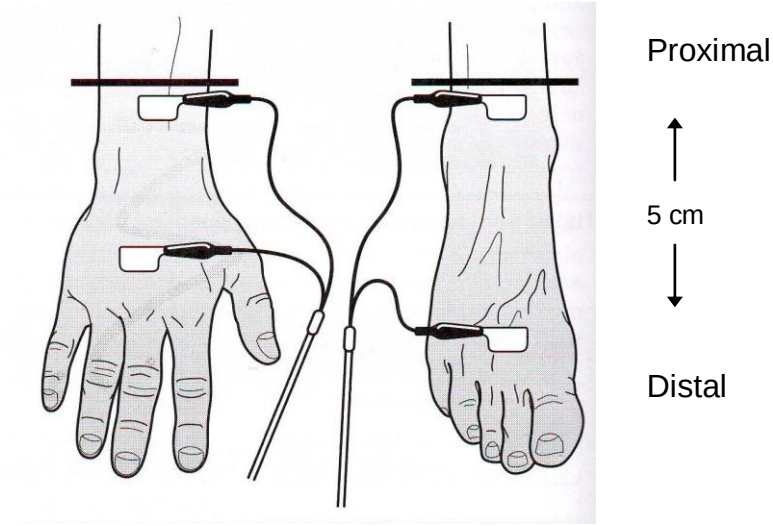


Figure 3. Proximal and distal electrode placement for whole-body BIA (taken from Haywood and Wagner, 2004)

These sites must be cleaned with an alcohol wipe and should be shaved to remove excess hair if required, in order to maximise conductivity between the electrodes and the surface of the skin. The participants arms and legs must be comfortably abducted (35-40°) is recommended (Heyward and Wagner, 2004), ensuring no contact between the thighs, and the arms and trunk. As this may “short circuit” the path of the electrical current, having a large affect on impedance value (Heyward and Wagner, 2004). Participant information is then manually entered into the device, including; height, weight, gender, age and activity level. When the subject is ready and has been in the supine position for precisely 10 minutes, the enter button is pressed to initiate measurement; an electrical current is sent through the body for approximately 5 seconds. After a beep is

sounded, results can be read from the digital display, at this point, electrodes may be removed from the participant.

Under water weighing (HW)

The participant is require to wear minimal clothing, i.e. tight swimsuit or Speedos along with a diving belt to stop the participant from floating to the surface of the tank, this is particularly important in more adipose individuals as excess fat will make the participant more buoyant. Before an under water measurement can be taken, the participant must have their weight out of water, and forced vital capacity (FVC) measured. FVC is measures using the Vitalograph Gold Standard. The weight is taken from the BodPod scales as they are regularly calibrated to optimise accuracy. These values are required as part of the calculation of body density, which is used to estimate %body fat. The participant is required to carefully enter the filled tank backwards facing the removable steps leading into the water, ensuring they keep the water calm upon entry, and avoid contact with the suspended seat attached to the load cell. The participant then submerges themselves fully under water, gently using their hands to eliminate any trapped air on the skin, hair and swimsuit. It is then recommended that the participant practices the technique of fully exhaling before submerging their head under water and attempting to expel any air left in the lungs whilst fully submerged. Once they are competent with this technique they are required to sit on the suspended chair, this may require familiarisation before the participant can balance on the seat. Once comfortable and balanced on the seat, the participant must repeat the

exhalation and submersion process whilst on the chair, avoiding contact with the sides of the tank, this is very important, so not to produce a false under- water weight. Once the participant is fully submersed on the seat and has fully expelled any remaining air to the best of their ability, they must try and remain still for as long as they comfortably can, to allow the under water weight to be recorded. As soon as the weight is recorded the experimenter will use verbal communication, instructing the participant to ascend from under the water so they are not holding their breath for longer than needed. This process is repeated until 3 readings are within 100g of each other (this usually takes 10 readings), the mean weight of these 3 values is used as the under water weight.

BodPod (ADP)

Participants are instructed to completely void their bowels and bladder before changing into a tight fitting swim suit and swimming cap. The participant height is measured to the nearest centimetre using a stadiometer. Then the participants information (participant ID number, height and gender) is entered into the accompanying computer, connected to the BodPod. The participants are instructed to remove jewellery and watches. At this point the software prompts the experimenter to conduct a two point calibration. This involves a baseline calibration of the empty chamber and phantom calibration using a 40.995-L metal calibration cylinder. Once calibration is complete the participant is prompted to step onto the BodPod scales. Whilst standing still, a weight measurement is acquired, the participant is then instructed to step off the

scales and enter the BodPod. The subject is shown the emergency stop button which releases the door locks if any problems occur. They are instructed to sit in the centre of the seat resting their back against the rear wall of the chamber with their feet in the centre of the base of the BodPod, legs apart and their hands on their lap. Before the door is gently closed they are instructed to remain still and breath normally. Once the door is shut the test can be initiated activating the magnetic door locks. The test last roughly 50 seconds, after the initial test the door is opened fully, the participant is asked if they are ready for a second test, if the participant is ready the door is gently shut and a second test is initiated (the amount of time the door is open between tests, should be standardised) . Once the tests are completed, the BodPod door is opened to avoid any sense of claustrophobia by the participant. If the two tests disagree by more than 150 ml, then a third test must be performed. If it remains that no two tests are in agreement by 150 ml, then the entire process including calibration is repeated until two tests meet this requirement. The two tests are then averaged and are used in the calculation of raw body volume. In order to calculate a body volume, height and weight must be used to estimate body surface area, predicted thoracic gas volume is also used in this calculation. Body surface area is required to account for the negative volume produced by the isothermal air surrounding the surface of the body. Thoracic gas volume (TGV) is also accounted for due to the isothermal air in the lungs and airway. Raw body volume, surface area artifact and TGV are used to produce a corrected body volume. This value is used to estimate body density which is then used to estimate %BF.

Appendix B: Informed research consent, PAR-Q and further declaration.

Informed research consent

A physiological research study for Ben Davies; Post graduate MSc by research student.

Please take time to carefully read the following informed consent document. When you have read and understood the information below, you will be invited to participate in the following research study. To confirm your participation you must accept the procedures outlined in this document.

The study centres on producing a more accurate estimation of body composition (i.e. % body fat and fat free mass) in Indian males, and is entitled:

Development of a scaling factor for more accurate estimation of Asian male adiposity from commercial methods.

The investigation requires you to have your body composition measured using the following methods: Air displacement plethysmography (ADP), bioelectrical impedance analysis (BIA), hydrostatic weighing (HW), skinfold analysis (SKF) and Magnetic resonance imaging (MRI). Participants are also required to conduct a forced vital capacity test (FVC) using the Vitalograph spirometry device. The investigation will take place at the sophisticated Sport Science laboratory of the University of Bedfordshire and the Steiner MRI unit at the Hammersmith hospital London. The investigation requires both Caucasian and Indian males
(specification of Indian participant: all four grandparents must be Indian).

All participants must be fasted from 4 hours prior to testing, participants should consume water as usual to maintain normal hydration.

Procedure for FVC- using the Vitalograph device:

Participants are required to have their height measured before testing, then the participant is required to take a full breath in, then exhale fully as much air as quickly and as powerfully as possible until they can no longer, through a disposable mouthpiece attached to the device. The subject will be seated as such exhalations can cause a short spell of light dizziness in some individuals.

Procedure for ADP-using the BodPod device:

The participant, wearing a tight fitting swimsuit and a swimming cap (provided by tester) has an accurate measure of body weight taken on the BodPod's accompanying scales. The participant then enters the chamber of the BodPod, sitting, with feet slightly apart, hands relaxed on the lap and their back straight, away from the machine wall. For an accurate measure the participant is required to breathe normally and sit very still, **it is extremely important the participant is wearing a tight fitting swimsuit.** This method requires two measures to be made simultaneously each lasting around 50 seconds.

Procedure for HW-using the under water weighing tank:

Wearing a tight fitting swimsuit and provided weight belt, the participant is required to enter the under water weighing tank which will contain water at a temperature of 30-35°C. The participant is required to be seated on the seat suspended from above the tank holding onto the chain of the seat for support. When the participant is ready they will exhale fully before submerging their head entirely under water, they must then expel as much of the remaining air in the lungs as possible. It is important the participant does not touch the sides of the tank and must remain still whilst submerged, to allow for an under water weight measurement to be taken. Once the tester has gained a weight measurement they will call down to the participant to bring their head out from under the water, if the participant feels the need to take a breath they should immediately bring their head out from the water. At no point should the participant feel pressured to hold their breath for longer than they are able. The technique will be repeated (possibly up to 10 times) until the tester has gained a consistent reading. The participant will have the opportunity to practice this technique until they are confident with it, beforehand; they will also observe the technique being administered on a volunteer to give a greater understanding of what is required. A robe will be provided for the participant to wear before and after entering the tank to avoid becoming cold.

Procedure for BIA-using the Tanita device:

The Participant, wearing a tight fitted swimsuit is required to step onto the Tanita device, placing both feet respectively onto the metal footplates on the base of the unit. Once the tester has entered the subject information into the device the participant is required to stand still in order to allow for a weight measurement to be acquired. They are then prompted to grasp the metal handles and hold them by their side. The device will then send a small current through the body for about 5 seconds, after a beep is sounded the participant can put the handles back on their mounts and step off the device.

BIA-using the Quadscan device:

This method requires the participant to be laid down on their back for 5 minutes to allow the fluid in the body to settle. During this period, the participant information is entered into the device. Then 4 adhesive electrodes are placed on the right side of the body, 2 on the hand, and 2 on the foot (each electrode site is to be cleaned using an alcohol wipe prior to attachment). The device then sends a small current down the right side of the body for about 10 seconds.

If you are fitted with a pacemaker it is unsafe to tested using any BIA methods and you will be unable to participate. As the electrical current my interfere with proper function of the pacemaker.

MRI and Skinfold analysis

You will be required to have measures taken at the Hammersmith hospital by John McCarthy, who is conducting a PhD research study, currently operating under existing ethical approval (ref. McCarthy, REC Ref. 06/Q0411/173). You will be subject to a whole body MRI scan lasting approximately 30 minutes, and you will also have your body composition estimated using skinfold calipers. You will be required to sign a separate informed consent prior to testing at the Hammersmith hospital.

Your safety will be assured, the researcher and technical support staff are competent users the apparatus detailed above.

You have the right not to participate. You can stop at any time if you do not want to finish the study, with no penalty. Your participation is not in response to any financial inducements.

All data will be anonymous, your data will be assigned an identification number, from then on your data will only be represented by this number. This prevents anyone else from knowing your results. All data will be stored in a locked folder that only the researcher and supervisor will have access to. At the end of the investigation, if you so wish, you can have access to your personal results.

If you are interested in receiving your results, or have any future questions, please contact me at **********@******.AC.UK

If you have any query regarding the above information, please ask now.
If you have read and understood these instructions, and you do not have any further questions, please sign below.

I agree to participate in the above study,

Signature of Participant: _____

Name of Participant (Block capitals): _____

Signature of Researcher: _____

Physical Activity Readiness Questionnaire - PAR-Q (Revised - July 2007)

PAR-Q & YOU

(A Questionnaire for People Aged 15 to 69)

Regular physical activity is fun and healthy, and increasingly more people are starting to become more active every day. Being more active is very safe for most people. However, some people should check with their doctor before they start becoming much more physically active.

If you are planning to become much more physically active than you are now, start by answering the seven questions in the box below. If you are between the ages of 15 and 69, the PAR-Q will tell you if you should check with your doctor before you start. If you are over 69 years of age, and you are not used to being very active, check with your doctor.

Common sense is your best guide when you answer these questions. Please read the questions carefully and answer each one honestly : Check YES or NO.

YES	NO	
☐	☐	Has your doctor ever said that you have a heart condition and that you should only do physical activity recommended by a doctor?
☐	☐	Do you feel pain in your chest when you do physical activity?
☐	☐	In the past month, have you had chest pain when you were not doing physical activity?
☐	☐	Do you lose your balance because of dizziness or do you ever lose consciousness?
☐	☐	Do you have a bone or joint problem (for example, back, knee or hip) that could be made worse by a change in your physical activity?
☐	☐	Is your doctor currently prescribing drugs (for example, water pills) for your blood pressure or heart condition?
☐	☐	Do you know of any other reason why you should not do physical activity?

If you answered

YES to one or more questions

Talk with your doctor by phone or in person BEFORE you start becoming much more physically active or BEFORE you have a fitness appraisal. Tell your doctor about the PAR-Q and which questions you answered YES.

- You may be able to do any activity you want - as long as you start slowly and build up gradually. Or, you may need to restrict your activities to those which are safe for you. Talk with your doctor about the kinds of activities you wish to participate in and follow his/her advice.
- Find out which community programs are safe and helpful for you.

NO to all questions

If you answered NO honestly to all PAR-Q questions, you can be reasonably sure that you can :

- start becoming much more physically active - begin slowly and build up gradually. This is the safest and easiest way to go.
- take part in a fitness appraisal - this is an excellent way to determine your basic fitness so that you can plan the best way for you to live actively. It is also highly recommended that you have your blood pressure evaluated. If your reading is over 144/94, talk with your doctor before you start becoming much more physically active.

➔

DELAY BECOMING MUCH MORE ACTIVE:

- if you are not feeling well because of a temporary illness such as a cold or a fever - wait until you feel better; or
- if you are or may be pregnant - talk to your doctor before you start becoming more active.

Please note: If your health changes so that you then answer "YES" to any of the above questions, tell your fitness or health professional. Ask whether you should change your physical activity plan.

Informed Use of the PAR-Q: The Canadian Society for Exercise Physiology, Health Canada, and their agents assume no liability for persons who undertake physical activity, and if in doubt after completing this questionnaire, consult your doctor prior to physical activity.

No changes permitted. You are encouraged to photocopy the PAR-Q but only if you use the entire form.

NOTE: If the PAR-Q is being given to a person before he or she participates in a physical activity program or a fitness appraisal, this section may be used for legal or administrative purposes.

"I have read, understood and completed this questionnaire. Any questions I had were answered to my full satisfaction."

Signature:_____ Identity Document No.:_____

Name:_____ Date:_____

Signature of Parent or Guardian:_____ Witness:_____
(for participants under the age of majority)

Note: 1.The information provided on this form will only be used for the application for use of Leisure and Cultural Services Department's Fitness Rooms and enrolment of recreation and sports activities. For correction of or access to personal data collected by means of this form, please contact staff of the enrollment counter/district.

2.If you answer "yes" to one or more questions in the "PAR-Q & YOU", your physical condition may not be suitable for taking part in the activity concerned. For safety's sake, you should consult a doctor in advance and produce a medical certificate upon enrolment or hire of fitness equipment to prove that you are physically fit for taking part in the activity. If you fail to produce a medical certificate, you must submit the completed Declaration upon enrolment or hire of fitness equipment.

This physical activity clearance is valid for a maximum of 12 months from the date it is completed and becomes invalid if your condition changes so that you would answer YES to any of the seven questions.

 Supported by: Health Canada

Decleration (please answer the following by circling the appropriate response)

1. **Are you fitted with a pacemaker?**

   Yes/No

2. **As far as you are aware, do you have an allergy to skin adhesive such as 'Elastoplast'?**

   Yes/No

3. **As far as you are aware, do you have an allergy to Savlon**

   Yes/No

Signature of applicant: _____

Name of applicant: _____

Date: _____

Appendix D: Body composition calculation equations of 2C methods and the Siri equation to estimate percent body fat.

BIA

-Unpublished.

Sum of four skinfolds (Durnin & Womersley, 1974)

-Sum of four skinfolds (mm) (bicep + tricep + subscapula + suprailliac).

-Body density = 1.1765 – 0.0744 × (LOG transformed sum of skinfolds)

HW

-Body density = body mass / {[(body mass - body mass in water) / density of

water at temperature when tested] – (residual volume + gastrointestinal)}

ADP

-Body volume (litres) = raw body volume – surface area artifact + 40% TGV.

-Body density = body mass / body volume.

Siri equation

-Percent body fat = (4.95 / body density – 4.50) × 100.

Appendix E: Assumptions of ANOVA- SPSS output.

Shaprio-Wilks

		Shapiro-Wilk		
	Ethnicity	Statistic	df	Sig.
Age	Caucasian	.875	11	.089
	Asian	.831	10	.034
Height	Caucasian	.947	11	.603
	Asian	.930	10	.449
Weight	Caucasian	.838	11	.030
	Asian	.908	10	.270
BMI	Caucasian	.892	11	.148
	Asian	.936	10	.505
MRI	Caucasian	.972	11	.905
	Asian	.951	10	.680
Tanita	Caucasian	.924	11	.350
	Asian	.950	7	.730
Bodystat	Caucasian	.846	11	.038
	Asian	.888	10	.159
HW	Caucasian	.933	11	.445
	Asian	.879	9	.154
BODPOD	Caucasian	.968	11	.867
	Asian	.970	10	.887
totalskf	Caucasian	.937	11	.490
	Asian	.849	10	.057
WHR	Caucasian	.893	11	.150
	Asian	.939	10	.543
Hip	Caucasian	.907	11	.226
	Asian	.908	10	.270
waist	Caucasian	.772	11	.004
	Asian	.920	10	.359
ViscPmass	Caucasian	.897	11	.168
	Asian	.979	10	.959
viscPadipose	Caucasian	.917	11	.295
	Asian	.939	10	.542
FVC	Caucasian	.930	11	.415

	Asian	.974	9	.930
Sub.abdoPadi	Caucasian	.949	11	.629
	Asian	.885	10	.150
subcutabdoPmass	Caucasian	.937	11	.483
	Asian	.817	10	.023
bicepskf	Caucasian	.697	11	.000
	Asian	.726	10	.002
tricepskf	Caucasian	.859	11	.055
	Asian	.852	10	.061
subscapskf	Caucasian	.932	11	.431
	Asian	.819	10	.025
illiaccskf	Caucasian	.807	11	.012
	Asian	.809	10	.019

Levene's test

Test of Homogeneity of Variance

		Levene Statistic	df1	df2	Sig.
Age	Based on Mean	13.139	1	19	.002
	Based on Median	11.223	1	19	.003
	Based on Median and with adjusted df	11.223	1	16.565	.004
	Based on trimmed mean	13.560	1	19	.002
Height	Based on Mean	3.867	1	19	.064
	Based on Median	3.013	1	19	.099
	Based on Median and with adjusted df	3.013	1	15.539	.102
	Based on trimmed mean	3.734	1	19	.068
Weight	Based on Mean	.116	1	19	.737
	Based on Median	.145	1	19	.707
	Based on Median and with adjusted df	.145	1	18.890	.707
	Based on trimmed mean	.168	1	19	.686
BMI	Based on Mean	.133	1	19	.719

	Based on Median	.115	1	19	.738
	Based on Median and with adjusted df	.115	1	18.928	.738
	Based on trimmed mean	.095	1	19	.761
MRI	Based on Mean	.244	1	19	.627
	Based on Median	.227	1	19	.639
	Based on Median and with adjusted df	.227	1	16.770	.640
	Based on trimmed mean	.231	1	19	.637
Tanita	Based on Mean	1.136	1	16	.302
	Based on Median	1.062	1	16	.318
	Based on Median and with adjusted df	1.062	1	13.668	.321
	Based on trimmed mean	1.064	1	16	.318
Bodystat	Based on Mean	.073	1	19	.789
	Based on Median	.069	1	19	.796
	Based on Median and with adjusted df	.069	1	18.978	.796
	Based on trimmed mean	.078	1	19	.783
HW	Based on Mean	.137	1	18	.716
	Based on Median	.190	1	18	.668
	Based on Median and with adjusted df	.190	1	17.953	.668
	Based on trimmed mean	.178	1	18	.678
BODPOD	Based on Mean	.943	1	19	.344
	Based on Median	.852	1	19	.368
	Based on Median and with adjusted df	.852	1	15.834	.370
	Based on trimmed mean	.979	1	19	.335
totalskf	Based on Mean	.367	1	19	.552
	Based on Median	.283	1	19	.601
	Based on Median and with adjusted df	.283	1	18.271	.601
	Based on trimmed mean	.379	1	19	.546
WHR	Based on Mean	4.541	1	19	.046
	Based on Median	3.775	1	19	.067

	Based on Median and with adjusted df	3.775	1	16.783	.069
	Based on trimmed mean	4.593	1	19	.045
Hip	Based on Mean	.746	1	19	.399
	Based on Median	.463	1	19	.504
	Based on Median and with adjusted df	.463	1	17.789	.505
	Based on trimmed mean	.713	1	19	.409
waist	Based on Mean	.639	1	19	.434
	Based on Median	.663	1	19	.426
	Based on Median and with adjusted df	.663	1	18.992	.426
	Based on trimmed mean	.658	1	19	.427
ViscPmass	Based on Mean	1.189	1	19	.289
	Based on Median	.307	1	19	.586
	Based on Median and with adjusted df	.307	1	16.360	.587
	Based on trimmed mean	1.069	1	19	.314
viscPadipose	Based on Mean	2.278	1	19	.148
	Based on Median	1.308	1	19	.267
	Based on Median and with adjusted df	1.308	1	14.968	.271
	Based on trimmed mean	2.260	1	19	.149
FVC	Based on Mean	.130	1	18	.723
	Based on Median	.145	1	18	.708
	Based on Median and with adjusted df	.145	1	17.319	.708
	Based on trimmed mean	.130	1	18	.723
Sub.abdoPadi	Based on Mean	.322	1	19	.577
	Based on Median	.262	1	19	.615
	Based on Median and with adjusted df	.262	1	17.533	.615
	Based on trimmed mean	.299	1	19	.591
subcutabdoPmass	Based on Mean	.005	1	19	.944
	Based on Median	.020	1	19	.888

	Based on Median and with adjusted df	.020	1	17.325	.889
	Based on trimmed mean	.000	1	19	.993
bicepskf	Based on Mean	1.224	1	19	.282
	Based on Median	.231	1	19	.636
	Based on Median and with adjusted df	.231	1	16.977	.637
	Based on trimmed mean	.944	1	19	.343
tricepskf	Based on Mean	.482	1	19	.496
	Based on Median	.257	1	19	.618
	Based on Median and with adjusted df	.257	1	18.870	.618
	Based on trimmed mean	.446	1	19	.512
subscapskf	Based on Mean	4.007	1	19	.060
	Based on Median	2.002	1	19	.173
	Based on Median and with adjusted df	2.002	1	11.475	.184
	Based on trimmed mean	3.477	1	19	.078
illiaccskf	Based on Mean	.593	1	19	.451
	Based on Median	.643	1	19	.432
	Based on Median and with adjusted df	.643	1	16.796	.434
	Based on trimmed mean	.520	1	19	.480

Mauchly's Test of Sphericity[b]

Measure:MEASURE_1							
Within Subjects Effect	Mauchly's W	Approx. Chi-Square	df	Sig.	Epsilon[a]		
					Greenhouse-Geisser	Huynh-Feldt	Lower-bound
bodyfat	.251	19.507	14	.152	.627	.846	.200

Tests the null hypothesis that the error covariance matrix of the orthonormalized transformed dependent variables is proportional to an identity matrix.

a. May be used to adjust the degrees of freedom for the averaged tests of significance. Corrected tests are displayed in the Tests of Within-Subjects Effects table.

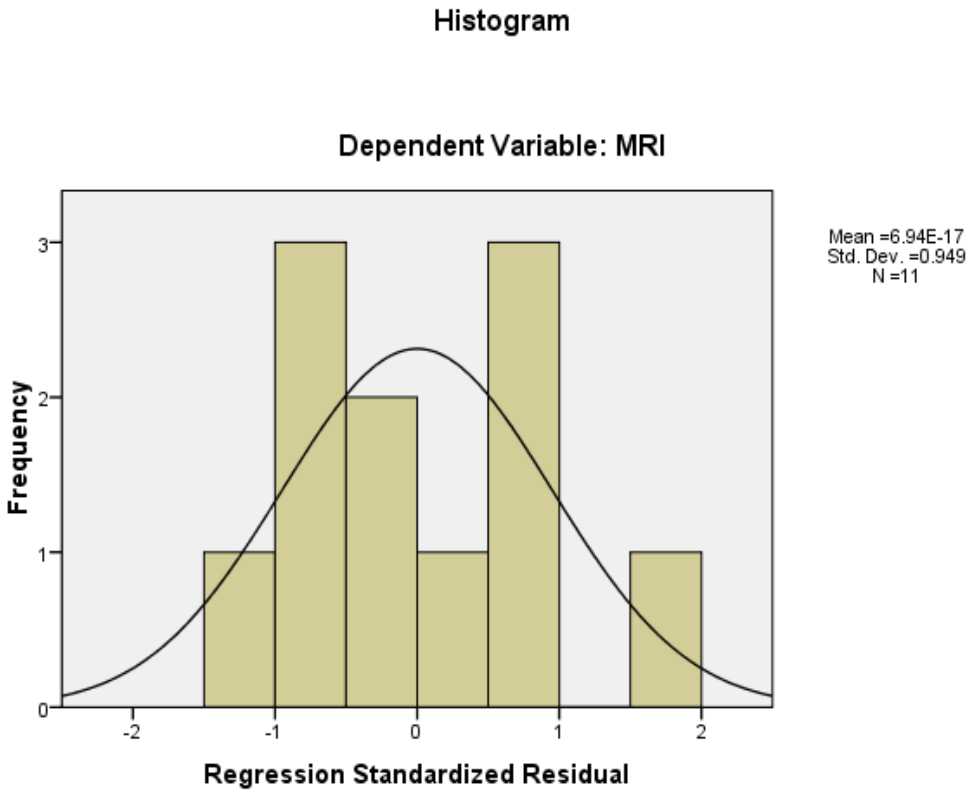
b. Design: Intercept + Ethnicity
Within Subjects Design: bodyfat

Mauchley's test of sphericity
Appendix F: Regression analysis-SPSS output.

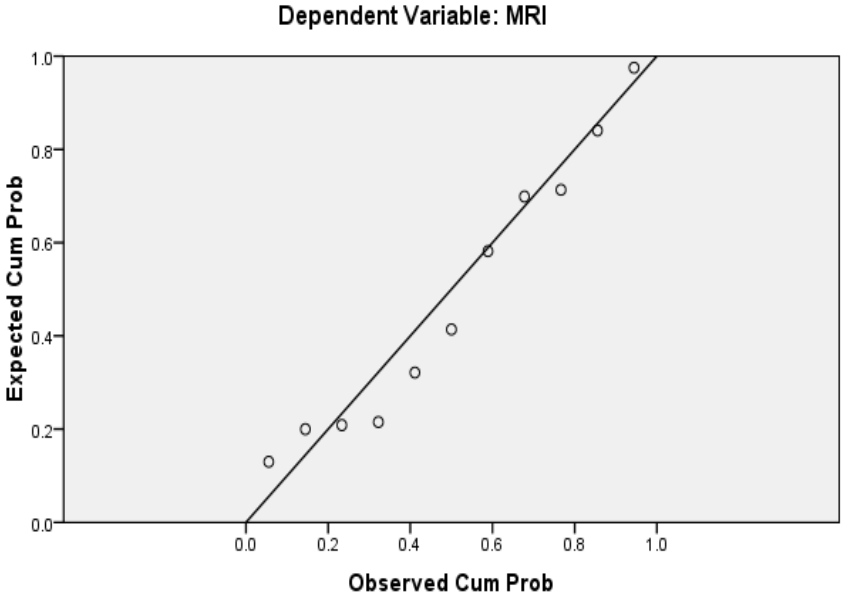
Caucasian regression

Descriptive Statistics

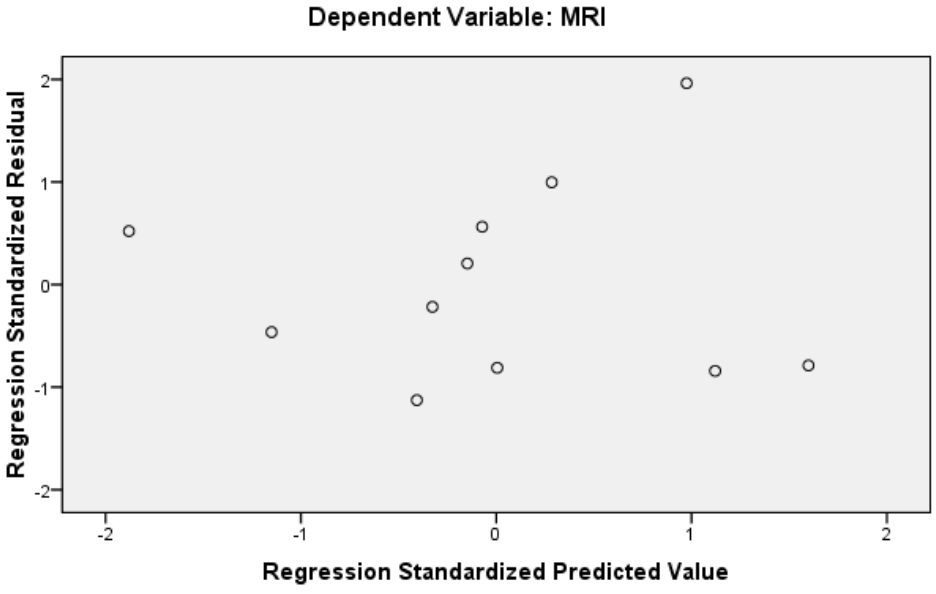
	Mean	Std. Deviation	N
MRI	17.1764	6.49281	11
Tanita	16.0818	5.70453	11
HW	18.9073	6.26201	11
totalskf	19.5733	5.42651	11
BODPOD	20.3609	7.53055	11
illiaccskf	19.8455	10.87671	11
abdomskf	21.6182	10.79331	11


Histogram
Dependent Variable: MRI
Mean =6.94E-17
Std. Dev. =0.949
N =11
Frequency
0
1
2
3
-2
-1
0
1
2
Regression Standardized Residual

Normal P-P Plot of Regression Standardized Residual



Scatterplot



Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	1.760	2.968		.593	.568					
	BODPOD	.757	.137	.878	5.508	.000	.878	.878	.878	1.000	1.000

a. Dependent Variable: MRI

Collinearity Diagnostics[a]

Model	Dimension	Eigenvalue	Condition Index	Variance Proportions	
				(Constant)	BODPOD
1	1	1.943	1.000	.03	.03
	2	.057	5.843	.97	.97

a. Dependent Variable: MRI

Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
					R Square Change	F Change	df1	df2	Sig. F Change
1	.878[a]	.771	.746	3.27376	.771	30.334	1	9	.000

a. Predictors: (Constant), BODPOD

b. Dependent Variable: MRI

ANOVA[b]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	325.109	1	325.109	30.334	.000[a]
	Residual	96.457	9	10.717		
	Total	421.566	10			

a. Predictors: (Constant), BODPOD

b. Dependent Variable: MRI

Coefficients[a]

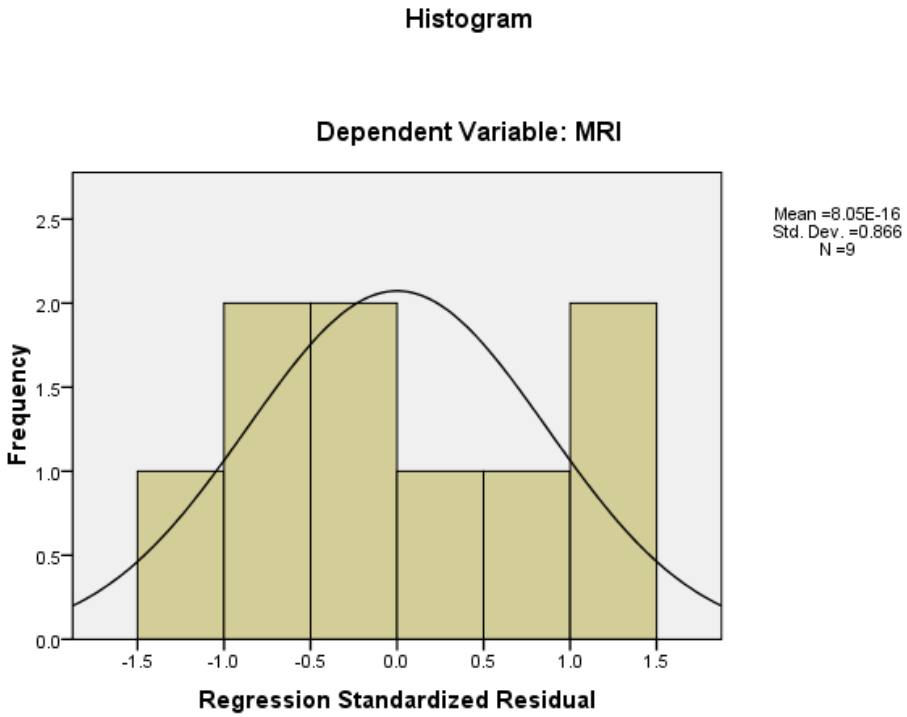
Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	1.760	2.968		.593	.568					
	BODPOD	.757	.137	.878	5.508	.000	.878	.878	.878	1.000	1.000

a. Dependent Variable: MRI

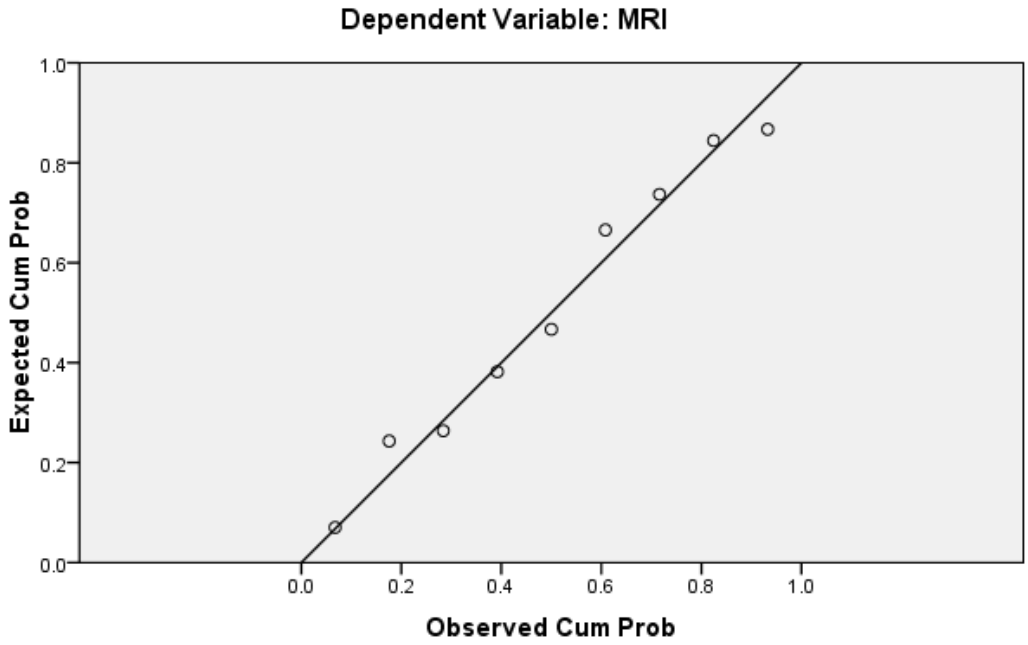
Asian Indian Regression

Descriptive Statistics

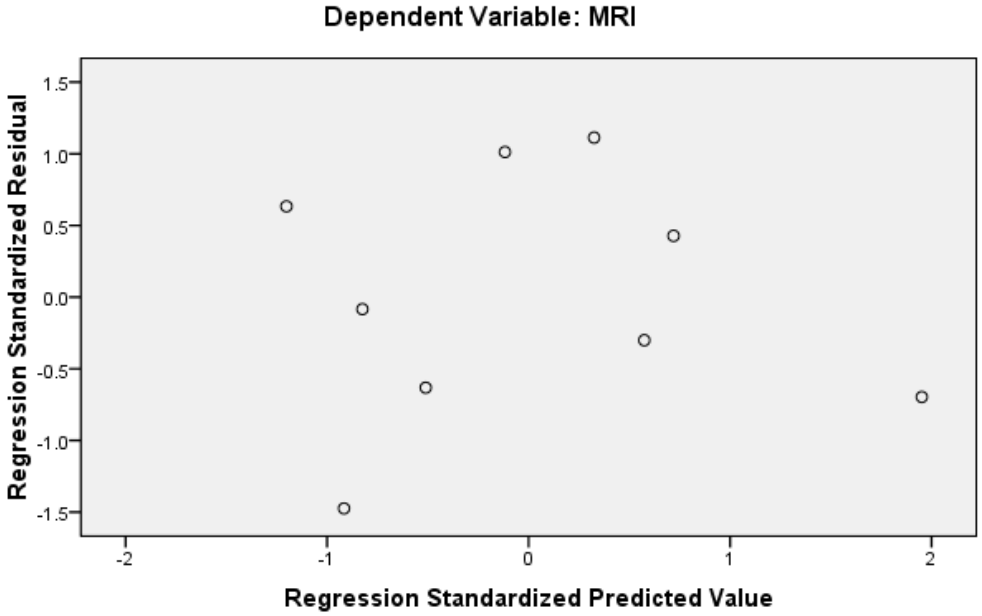
	Mean	Std. Deviation	N
MRI	30.1856 %	5.55628	9
Bodystat	19.9556 %	4.58151	9
HW	26.1322 %	5.68721	9
totalskf	26.8339 %	4.57741	9
BODPOD	30.9533 %	5.19491	9
WHR	.8933	.05657	9
waist	87.5333 cm	10.30801	9
bicepskf	7.7556 mm	3.68413	9
tricepskf	13.8889 mm	5.11333	9
subscapskf	23.3222 mm	10.53207	9



Normal P-P Plot of Regression Standardized Residual



Scatterplot



Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients			Correlations			Collinearity Statistics	
		B	Std. Error	Beta	t	Sig.	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	17.327	3.161		5.481	.001					
	tricepskf	.926	.215	.852	4.306	.004	.852	.852	.852	1.000	1.000
2	(Constant)	7.812	2.963		2.636	.039					
	tricepskf	.716	.132	.659	5.435	.002	.852	.912	.605	.843	1.186
	HW	.476	.118	.487	4.014	.007	.748	.854	.447	.843	1.186

a. Dependent Variable: MRI

Collinearity Diagnostics[a]

Model	Dimension	Eigenvalue	Condition Index	Variance Proportions		
				(Constant)	tricepskf	HW
1	1	1.945	1.000	.03	.03	
	2	.055	5.931	.97	.97	
2	1	2.917	1.000	.00	.01	.00
	2	.063	6.824	.14	.96	.06
	3	.020	12.068	.86	.03	.93

a. Dependent Variable: MRI

Model Summary[c]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
					R Square Change	F Change	df1	df2	Sig. F Change
1	.852[a]	.726	.687	3.10954	.726	18.543	1	7	.004
2	.962[b]	.926	.901	1.74956	.200	16.112	1	6	.007

a. Predictors: (Constant), tricepskf

b. Predictors: (Constant), tricepskf, HW

c. Dependent Variable: MRI

ANOVA[c]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	179.293	1	179.293	18.543	.004[a]
	Residual	67.685	7	9.669		
	Total	246.978	8			
2	Regression	228.612	2	114.306	37.343	.000[b]
	Residual	18.366	6	3.061		
	Total	246.978	8			

a. Predictors: (Constant), tricepskf

b. Predictors: (Constant), tricepskf, HW

c. Dependent Variable: MRI

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	17.327	3.161		5.481	.001					
	tricepskf	.926	.215	.852	4.306	.004	.852	.852	.852	1.000	1.00
2	(Constant)	7.812	2.963		2.636	.039					
	tricepskf	.716	.132	.659	5.435	.002	.852	.912	.605	.843	1.18
	HW	.476	.118	.487	4.014	.007	.748	.854	.447	.843	1.18

a. Dependent Variable: MRI

Appendix G: Power calculations (Cohen 1989)

$$n = (2(SD)^2*(Z_\alpha+Z_\beta))/\Delta^2$$

where, SD = standard deviation of the differences between sample groups

Z_α = Z-coefficient for the false-change (Type I) error rate from the table below.

Z_β = Z-coefficient for the missed-change (Type II) error rate from the table below.

Δ^2 = Minimum detectable change size. This needs to be specified in absolute terms rather than as a percentage.

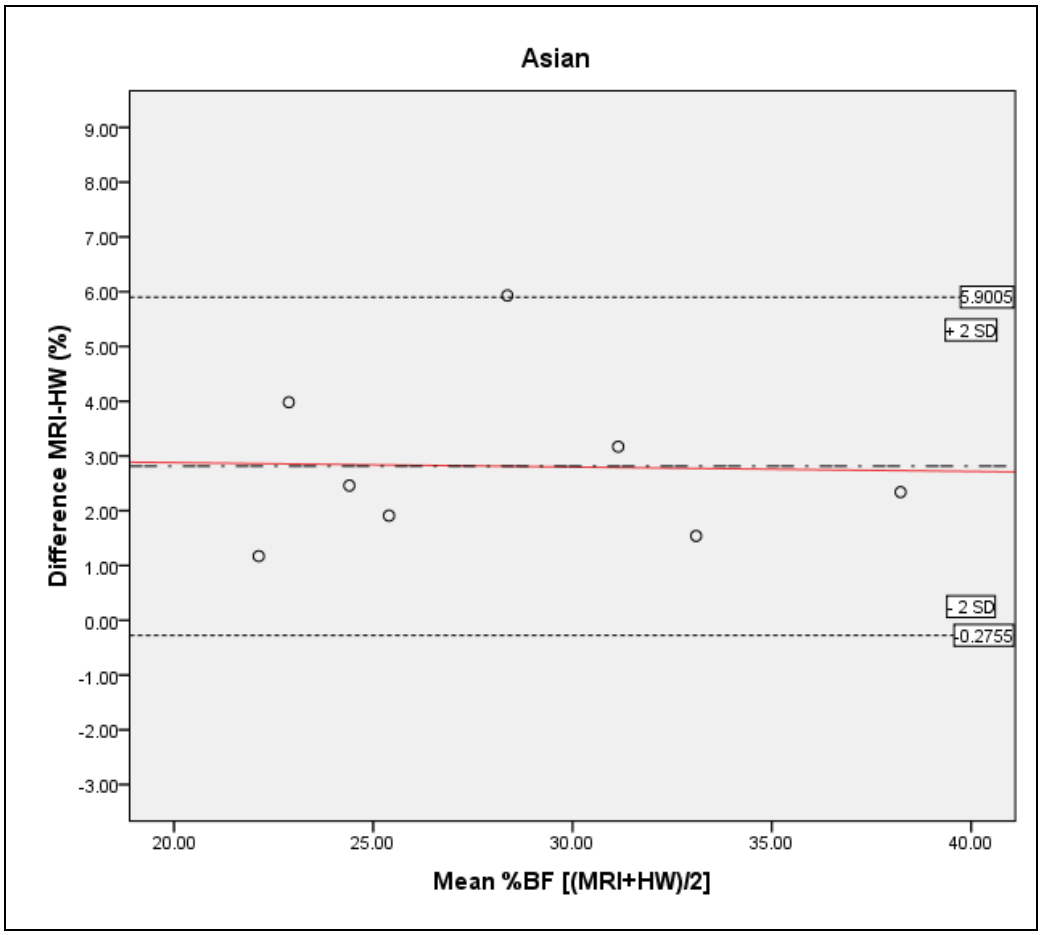
For example, if you wanted to detect a 20% change in the sample mean from one year to the next and your first year sample mean = 10 plants/quadrat then MDC = (0.20 x 10) = 2 plants/quadrat.

Table of standard normal deviates for $Z_{æ}$		Table of standard normal deviates for $Z_{ß}$		
False-change (Type I) error rate (æ)	$Z_{æ}$	Missed-change (Type II) error rate (ß)	Power	$Z_{ß}$
0.4	0.84	0.4	0.6	0.25
0.2	1.28	0.2	0.8	**0.84**
0.1	1.64	0.1	0.9	1.28
0.05	**1.96**	0.05	0.95	1.64
0.01	2.58	0.01	0.99	2.33

Mean Grp 1	**29.905**	asian	mri
Mean Grp 2	**31.018**	asian	adp
SD (Grp 2 - Grp 1)	**-0.41102**	5.3131	4.90208
MDC =	**0.01**		
n =	**10.2**		

Mean Grp 1	**17.1764**	cauc	Mri
Mean Grp 2	**20.3609**	cauc	Adp
SD (Grp 2 - Grp 1)	**1.03774**	6.49281	7.53055
MDC =	**0.01**		
n =	**171.2**		

Appendix H: Bland-Altman plot (MRI-HW) with outlier removed



Bland and Altman plot showing bias of agreement between MRI and HW percent body fat values (difference between MRI and HW against their mean) with outlying data point remaoved. Central line is group mean difference and the outer lines represent ± 2 standard deviations.

Appendix I:

UNIVERSITY OF BEDFORDSHIRE

Research Ethics Scrutiny (Annex to RS1 form)

Candidate: Ben Davies

Registration No.: 04269482

Research Institute: The School of PE and Sports Science

Research Topic: Development of a scaling factor for more accurate estimation of Asian male adiposity from commercial methods.

SECTION A to be completed by the candidate

The candidate is required to summarise in the box below the ethical issues involved in the research proposal and how they will be addressed. In any proposal involving human participants the following should be provided:

- clear explanation of how informed consent will be obtained,
- how will confidentiality and anonymity be observed,
- how will the nature of the research, its purpose and the means of dissemination of the outcomes be communicated to participants,
- how personal data will be stored and secured
- if participants are being placed under any form of stress (physical or mental) identify what steps are being taken to minimise risk

If protocols are being used that have already received UREC ethical approval then please specify. Roles of any collaborating institutions should be clearly identified. Reference should be made to the appropriate professional body code of practice.

Answer the following question by ringing/deleting yes or **no** as appropriate:

1. Does the study involve vulnerable participants or those unable to give informed consent (e.g. children, people with learning disabilities, your own students)?
   No

2. Will the study require permission of a gatekeeper for access to participants (e.g. schools, self-help groups, residential homes)?
   No

3. Will it be necessary for participants to be involved without consent (e.g. covert observation in non-public places)?
   No

4. Will the study involve sensitive topics (e.g. sexual activity, substance abuse)?
   Yes
5. Will blood or tissue samples be taken from participants?
   No

6. Will the research involve intrusive interventions (e.g. drugs, hypnosis, physical exercise)? **Yes (immersion for underwater weighing)**

7. Will financial or other inducements be offered to participants (except reasonable expenses)? **No**

8. Will the research investigate any aspect of illegal activity? **No**

9. Will participants be stressed beyond what is normal for them? **Yes (underwater weighing)**

10. Will the study involve participants from the NHS (e.g. patients or staff)? **No**

If you have answered yes to any of the above questions or if you consider that there are other significant ethical issues then details should be included in your summary above. If you have answered yes to Question 1 then a clear justification for the importance of the research must be provided.

*Please note if the answer to Question 10 is yes then the proposal should be submitted through **NHS research ethics approval procedures** to the appropriate **COREC**. The University Research Ethics Committee should be informed of the outcome

Prior to any testing, participants will be briefed and made fully aware of the aims and procedures of the investigation by reading a clear and detailed informed consent form, which they will sign to confirm their participation in the study (see Appendix A). Participants will also be medically screened (see Appendix B) to ensure their state of health is adequate for participation.

All participants will be assigned an identification number that will anonymise their identity; only the researcher will hold details of the identity of the participants. All personal data will be stored in a secure file that only the researcher and their supervisor has access to. Participants will be made aware that their information will not be used beyond the scope of the immediate research setting and that there will be no way of identifying an individual participant from their data. Participants will be de-briefed after testing and will have private access to their own data if they so wish upon completion of the investigation.

The topic of body fat can be a sensitive one as some individuals may feel uncomfortable about their body composition and health. Participants will be fully assured of their anonymity during the entire investigation and will also be made aware that they are required to wear minimal clothing in the form of a tight swim suit; it is important to ensure the privacy of the participant whilst they are wearing such attire. Therefore, participants will get changed in a separate, locked changing room in which a robe will be made available to cover their body. They will only be requested to remove the robe when appropriate - immediately prior to being tested. They will have access to a privacy screen for disrobing and approach to the immersion tank and the BodPod if they wish. Only the researcher and any necessary technical support staff will be in the room with the participants. Technical support will be required for safety during under water weighing.

Before entering the underwater weighing tank the participant will observe a volunteer who will perform a demonstration of the technique. This is done to increase awareness of the procedure prior to being tested. The under water weighing procedure requires the participant to exhale fully before submerging entirely under water. They are then required to expel any remaining air in the lungs. This is very important for accurate testing. This may prove an unnatural and possibly uncomfortable experience. In order to improve the experience, participants will be slowly introduced to the tank and will practice the immersion/exhalation technique until they are happy to continue. Participants will remain under water holding their breath for as long as they are comfortably able until they feel

the need to take a breath. At no point will the participant be pressured to hold their breath for longer than they are comfortable. As soon as the researcher has gained a reading they will inform the participant immediately in order to avoid the participant holding their breath for longer than required.

Manufacturers of bioelectrical impedance devices warn not to allow those with pacemakers to be analysed using bioelectrical methods. It will therefore be important to identify any participants fitted with a pacemaker as the bioelectrical impedance device sends an electrical current through the body which may interfere with proper functioning of the pacemaker.

The Bioelectrical impedance devices requires the use of an adhesive electrode placed upon the skin of the hands and feet. Therefore, if the participant is aware of any allergy to skin adhesive (for example 'Elastoplast' and similar) they will be excluded form testing with this machine.

No other methods or devices proposed for use in this investigation have been identified as a potential cause of physical or mental harm.

Checklist of documents which should be included:

- Project proposal (with details of methodology) & source of funding
- Documentation seeking informed consent (if appropriate)
- Information sheet for participants (if appropriate)
- Questionnaire (if appropriate)

Signature of Applicant: Date: 20/11/08

Signature of Director of Studies: Date: 20/11/08

This form together with a copy of the research proposal should be submitted to the Research Institute Director for consideration by the Research Institute Ethics Committee/Panel

Note you cannot commence collection of research data until this form has been approved

SECTION B Consideration by Research Institute Ethics Committee/Panel

Comments:

Approved

Signature Chair of Research Institute Ethics Committee/Panel:

Date:

This form should then be filed with the RS1 form

If in the judgement of the committee/panel there are significant ethical issues for which there is not agreed practice then further ethical consideration is required before approval can be given and the proposal with the committee/panel's comments should be forwarded to the secretary of the UREC for consideration.

There are significant ethical issues which require further guidance

Signature Chair of Research Institute Ethics Committee/Panel:

Date:

This form together with the recommendation and a copy of the research proposal should then be submitted to the University Research Ethics Committee

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