2012

Validity of non-invasive methods for body composition measurements in older adults

Yulong Li
Iowa State University

Recommended Citation

Li, Yulong, "Validity of non-invasive methods for body composition measurements in older adults" (2012). *Graduate Theses and Dissertations*. 12598.
https://lib.dr.iastate.edu/etd/12598

Validity of non-invasive methods for body composition measurements in older adults

by

Yulong Li

A thesis submitted to the graduate faculty

in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

Major: Kinesiology

Program of Study Committee:

Rick L. Sharp, Major Professor

Tim R. Derrick

Warren D. Franke

Gregory J. Welk

Iowa State University

Ames, Iowa

2012

Table of Contents

ACKNOWLEDGEMENTS

I would very much like to thank my major professor, Dr. Rick Sharp, for his great guidance, support and encouragement throughout my master's study. Great thanks to Dr. Tim Derrick, Dr. Warren Franke and Dr. Gregory Welk for their time and effort contributed to this project.

I also wanted to express my gratitude to all of the faculties and students who helped with data collection in South Dakota State University. I appreciate the help of research associates Hector Angus and Jean Stewart for their effort in keeping everything in order and happen in time. Huge thanks to my great friends from the department, Yang Bai, Youngwon Kim and Jungmin Lee, who generously offered great help to my data analysis and writing, I couldn't have finished all this without their help. Thanks to peer master students Andrea Banks, Katelin Blasingame, Christina Sauder and Benjamin Wetli for the wonderful two years they brought to my life.

Finally, I would like to thank my great parents for their constant love and support throughout my study here in the United States, and my boyfriend, Yongzhang Bai for always being there for me even though we have been overseas apart.

ABSTRACT

Purpose: The aim of this study was to investigate the validity of bioelectrical impedance analysis (BIA) and air-displacement plethysmography (ADP) in relation to dual energy X-ray absorptiometry (DXA) (i.e. the reference standard). **Methods:** Sixty-three older adults aged 60-96 years (40 men, 23 women). Body percent fat (%BF) was estimated by BIA, ADP and DXA. Single frequency (50Hz) BIA that measures whole body impedance was used and Kyle's equation was applied to estimate fat-free mass. Paired sample t-test, absolute percent errors and Cohen's d were used to evaluate differences among the 3 different methods. Methods agreement was assessed by Pearson's correlation, regression analysis and Bland-Altman plots. Classification agreement of obesity was evaluated using Kappa statistics. **Results:** ADP and BIA significantly overestimated %BF relative to DXA by 3.3% and 3.1%, yielding absolute errors of 14.1% and 12.4%, respectively. However, ADP (Cohen's d=0.35) had better agreement with DXA and BIA (Cohen's d=0.40). Regression analysis indicated smaller individual variations of ADP (SEE=3.23) compared to BIA (SEE=4.78). In addition, ADP (Kappa=0.58) showed better obesity classification agreement relative to DXA in comparison with BIA (Kappa = 0.35). However, Bland-Altman plots showed a positive proportional pattern (Slope= 0.24, R^2=0.24, $p<0.05$) of biases in ADP, while no systematic pattern of biases was observed for BIA. A gender difference was also detected, indicating a better agreement in males than females. **Conclusion:** Given that both BIA and ADP had acceptable agreement with DXA in estimating %BF of older adults, ADP showed relatively better agreement in body composition measurement (i.e. %BF) and obesity classification in

comparison to BIA. However, practitioners and/or researchers should be aware of the potential biases when using ADP to estimate %BF in older populations.

CHAPTER 1 INTRODUCTION

Age-dependent loss of muscle mass and strength is growing to be a major cause of disability and morbidity among older adults (Roubenoff & Hughes, 2000). These alternations impair physical functional status, quality of life and increase the risk of comorbidities (Baumgartner, 2000). While exercise and nutritional interventions have been proved to be effective to offset these alternations (Benton, 2011; Nissen et al., 1996; Panton, Rathmacher, Baier, & Nissen, 2000; Thomson, 2009), an accurate estimation of body composition is essential to assess the changes that happen along with the interventions.

Although hydrodensitometry (HW) is a well-established reference standard in the assessment of body composition, it has many flaws that keep it from being a practical technique for participants. For example, the repeated weighing under water (Dempster & Aitkens, 1995) requires a great amount of cooperation of participants with practitioners/researchers. Also, any air remaining in the lungs during submersion will affect the measurement of lung volume, which influences the validity of the measurement (Wagner & Heyward, 1999).

Dual energy x-ray absorptiometry (DXA) is another reference method that yields precise measurement of body composition (Kiebzak, Leamy, Pierson, Nord, & Zhang, 2000) obesity (Pietrobelli, Formica, Wang, & Heymsfield, 1996; L. D. Plank, 2005; Prior, Cureton, Modlesky, Evans, & Sloniger, 1997). Since DXA is minimally stressful for most individuals (Ballard, Fafara, & Vukovich, 2004), it has been widely used as a reference standard in previous studies (S. Ball & Altena, 2004; Koda, Tsuzuku, Ando, Niino, & Shimokata, 2000;

L. Plank, 2005; Prior et al., 1997; Ravaglia et al., 1999; Sardinha, Lohman, Teixeira, Guedes, & Going, 1998; G. Sun, 2005; Wagner, Heyward, & Gibson, 2000).

Air displacement plethysmography (ADP), known as Bod Pod (Life Measurement Instruments, Concord, CA), has been a validated laboratory method for body composition measurement in different populations including children (Holmes, Gibson, Cremades, & Mier, 2011), adults (S. D. Ball, 2004; Ballard, et al., 2004; Wagner, et al., 2000), older adults (Sardinha, 1998, Bosy-Westphal 2003, Koda 2000) and athletes (Ballard, et al., 2004). Bod Pod predicts body composition by measuring body volume in a two-chamber structure (Dempster & Aitkens, 1995). It is comfortable, convenient and does not require high techniques to perform (Bosy-Westphal, 2003). In numerous studies that validated ADP against criteria measurements (i.e., HW, DXA) for adults (Aleman-Mateo et al., 2007; S. D. Ball, 2004; Bosy-Westphal, 2003; Ginde, 2005; Koda, et al., 2000; Sardinha, et al., 1998; Wagner, et al., 2000; Yee et al., 2001), its reported validity has not been consistent and conclusive. In support, some studies showed good agreement and/or validity of ADP against reference standards such as DXA and HW (Aleman-Mateo, et al., 2007; S. D. Ball, 2004; Ginde, 2005; Koda, et al., 2000; Yee, et al., 2001). In contrast, other studies indicated either overestimation (S. D. Ball, 2004; Bosy-Westphal, 2003; Wagner, et al., 2000) or underestimation (Sardinha, et al., 1998) of percent body fat (%BF). A few studies investigated the validity of ADP in older adults (Aleman-Mateo, et al., 2007; Bosy-Westphal, 2003; Koda, et al., 2000; Yee, et al., 2001) and the results varied by the different criterion measurements used.

Bioelectrical impedance analysis (BIA) has been used as a field method for body composition measurement due to its non-invasiveness, easiness to use, and relatively low cost. BIA measures %BF by measuring the resistance to an electric current that is introduced to the human body (Kushner, 1992). Fat free mass (FFM) is differentiated from fat mass (FM) in that it has high water and mineral content and is more conductive than FM (Kushner, 1992). Studies that compared BIA to reference methods (i.e., underwater weighing, DXA) in different populations showed contradictory results (Company & Stephen, 2010; Huygens, Claessens, Thomis, Loos, & Van Langendonck, 2002; Jaffrin & Morel, 2008; Savastano, 2010; Shafer, 2009; G. Sun, 2005) mainly due to variations in hydration status and the arbitrary choice of predictive equations to calculate %BF (Genton et al., 2001; Kyle et al., 2004). For older adults, since body composition and hydration status change along with aging (Waki, 1991), whether BIA could accurately measure body composition remains unanswered.

Obesity is associated with a higher prevalence of cardiovascular disease, metabolic disease, several important cancers, and numerous other medical conditions (Samper Ternent & Al Snih, 2012). While many physical activity/nutritional intervention programs have been developed, it is essential to categorically identify obese from non-obese when making decisions regarding intervention as well as follow-up policies (Himes, 1999). Therefore, it is critical that measurement tools for body composition provide precise estimation in classifying obese from non-obese populations.

The purpose of this study was to evaluate the validity as well as classification agreement of ADP and BIA in body composition measurement among older adults in relation to DXA as a reference method. It was hypothesized that both BIA and Bod Pod would serve as valid methods to measure FFM and %BF.

CHAPTER 2 REVIEW OF LITERATURE

The assessment of body composition

An accurate assessment of body composition is necessary to properly identify an individual's health risk associated with an excessively low or high body fat (%BF) (Wagner & Heyward, 1999). Periodic body composition measurements can help assess the change of %BF that happens along with any exercise/nutrition interventions or weight loss programs.

Numerous tools and methodologies have been developed to measure body composition including laboratory methods and field methods. Four commonly used laboratory methods include hydrodensitometry (underwater weighing), air displacement plethysmography (Bod Pod, Life Measurement Instruments, Concord, CA), isotope dilution, and dual-energy X-ray absorptiometry (DXA). Field methods include bioelectrical impedance analysis (BIA), near-infrared interactance (NIR), skinfolds (SKF), and anthropometric circumference measurement (Wagner & Heyward, 1999). However, even though these measurements tools have been validated and used in young adults (Laurson, Eisenmann, & Welk, 2011; Leahy, O'Neill, Sohun, & Jakeman, 2012; Panotopoulos, Charles, Bernard, & Arnaud, 2001; Tseh, Caputo, & Keefer, 2010), it is questionable whether they would serve as valid methods for older adults.

Underwater weighing has been considered the 'gold standard' for body composition measurement. It measures body volume by applying Archimedes' Principle, which states that the volume of an object is equal to the object's loss of weight in water with appropriate correction for the density of the water. However, since it requires repeated measures of the air present in the lungs during submersion, any air remaining in the lungs can result in large measurement errors (Dempster & Aitkens, 1995). In addition, it requires considerable amounts of training for technicians. Therefore, despite the high accuracy in measuring body composition, underwater weighing may not be practical, especially for older adults or people with physical disabilities who may have problems with submerging (Wagner & Heyward, 1999).

Dual energy x-ray absorptiometry (DXA) is a three-component model which assumes that the body consists of three components that are distinguishable by their X-ray attenuation properties, namely bone mineral content (BMC), fat mass (FM), and fat free mass (FFM) (L. Plank, 2005). The measurement involves the participant lying supine on an open table with an X-ray beam passing through the bone and soft tissue upward to a detector (Mazess, Barden, Biesek, & Hanson, 1990). The open table is minimally stressful for most individuals during measurement (Ballard, et al., 2004). As far as accuracy, although it has been reported that DXA significantly underestimated %BF in athletes (Arngrimsson, 2000), it has a good performance with healthy adults. For example, Prior et al. suggested no significant difference in %BF measurement between DXA and 4C model in healthy adults (p=0.10, (Prior, Cureton, Modlesky, Evans, Sloniger, et al., 1997)). In addition, DXA is not likely to be affected by factors that may potentially influence body composition measurement. For example, hydration changes have little effect on DXA estimates. For instance, a change of 1kg in extracellular fluid induces an estimation error of only 0.6% fat, which is less than one half the error associated with underwater weighing for the same change in body hydration status (Pietrobelli, 1996). Moreover, DXA is not affected by race, athletic status, or musculoskeletal development (Prior, Cureton, Modlesky, Evans, Sloniger, et al., 1997), and it requires minimal cooperation from the participant. Therefore, DXA is considered the reference standard for older adults.

Skinfolds have been widely used as a field method in body composition measurement due to its practicality (i.e., quick and non-invasive). However, for older adults, skinfold thickness may lead to errors in body composition prediction because it does not measure the increase of intra-abdominal fat mass that occurs with age (Han, Carter, Currall, & Lean, 1996).

Therefore, accurate and feasible measurements of body composition in older adults are essential. ADP and BIA are feasible options for older adults because they are quick, easy to perform, and less invasive. Their usage and validity will be discussed in the following sections.

Principles of air-displacement plethysmography

Air-displacement plethysmography – known as Bod Pod (Life Measurement Instruments, Concord, CA) - has been available commercially since its introduction in the market in 1995. It is a quick, comfortable, automated, non-invasive and safe method that can be accommodated to various participants (i.e., children, obese, elderly, and people with disabilities) (Fields, Goran, & McCrory, 2002).

Bod Pod consists of a single structure containing two chambers (front and rear), with a volume-perturbing element in the form of a moving diaphragm mounted in the middle. During operation, volume change in the two chambers produces pressure change, leading to oscillation of the electronically controlled diaphragm that records volumetric change of the chambers (Dempster & Aitkens, 1995).

A complete body composition test using Bod Pod involves measurement of uncorrected body volume, computation of the surface area artifact, and measurement of thoracic gas volume (V_{TG}). The subjects sit motionlessly in the front chamber with minimal clothing and breathe normally. Compressing clothes such as bathing suits and swimming caps are used to maintain adiabatic conditions. Body volume and density are determined using the following equations (Dempster & Aitkens, 1995):

$$\text{Body Volume (L)} = \text{Body Volume}_{raw} - \text{Surface Area Artifact} + 40\%V_{TG}$$

$$\text{Body Surface Area (cm}^2\text{)} = 71.84 * \text{Weight (kg)}^{0.425} * \text{Height (cm)}^{0.725}$$

*Surface Area Artifact = k (L/cm^2) * BSA (cm^2), where k is a negative constant derived by the manufacturer.*

$$V_{TG}\text{ (L)} = \text{Functional Residual Capacity} + 0.5\text{Tidal Volume}$$

$$\text{Functional Residual Capacity (L)} = 0.0472 \text{ (height in cm)} + 0.0090 \text{ (age)} - 5.290 \text{ (women)}$$

$$\text{Functional Residual Capacity (L)} = 0.0360 \text{ (height in cm)} + 0.0031 \text{ (age)} - 3.182 \text{ (men)}$$

Body Density (kg/L) = Body Weight/Body Volume

Validity of air-displacement plethysmography (Bod Pod)

A summary of studies that compared body-composition measurements by the Bod Pod and DXA in adults is shown in Table 1. Most of these studies were conducted in young to middle aged subjects (S. D. Ball, 2004; Ballard, et al., 2004; Sardinha, et al., 1998; Wagner, et al., 2000), including two studies conducted in older adults aged around or over 60yrs (Bosy-Westphal, 2003; Koda, et al., 2000).

BMI ranged from 19 to 30 for most participants involved. Of the 4 studies conducted in adults, only one study (Ballard, et al., 2004) showed no difference between Bod Pod and DXA. Differences between Bod Pod and DXA in %BF measurement ranged from -3.7% to 2.1% in the other 3 studies. One study suggested that Bod Pod underestimated %BF (Sardinha, et al., 1998), while others reported overestimation in %BF with Bod Pod (S. D. Ball, 2004; Wagner, et al., 2000). Two studies (S. D. Ball, 2004; Wagner, et al., 2000) reported correlation coefficients, and showed a significant correlation between Bod Pod and DXA in %BF measurement. One study (Ballard, et al., 2004) that conducted regression analyses showed a significant linear relation between ADP and DXA, with slopes of 1.1-1.2 (R^2=0.83-0.85; SEE=2.14-2.83).

Both studies that were conducted in older adults (Bosy-Westphal, 2003; Koda, et al., 2000) involved subjects aged 40-82 yrs (mostly over 60 yrs). They both indicated significant correlations in %BF measured by Bod Pod and DXA. In one study

(Bosy-Westphal, 2003), Bod Pod significantly overestimated %BF than DXA, while in the other (Koda, et al., 2000), the results varied depending on age, sex and BMC.

Table 1. Summary of studies that compared ADP to DXA

							Regression analysis			Bland-Altman analysis
Reference	**N**	**Sex**	**Age**	**BMI**	**%BF (ADP-DXA)**	**r**	**Slope**	**R^2**	**SEE**	**95%LoA[c]**
Adults										
Ball	160	M	32.0±11.0	23.0 ± 4.1	2.1±3.0**	0.94**	NR	0.89	NR	NR
Ballard	47	F	19.7 ± 1.0	22.7 ± 2.3	0	NR	1.097	0.85	2.14	NR
	24	F	20.0 ± 1.5	22.8 ± 3.5	0	NR	1.166	0.83	2.83	NR
Wagner	30	M	32.0 ± 7.7	25.9 ± 4.2	1.7 **	0.91**[1]	NR	0.86	2.84	NR
Sardinha	62	M	37.6 ± 2.9	27.8 ± 3.5	-2.6±2.7*	0.93	NR	0.90	2.4	-2.6, 7.8
Older										
Bosy-Westphal	26	M,F	67.7±6.6	26.4±3.2	1.6 ±3.1**	NR	0.85 **	0.87	NR	6.8
Koda	721	M,F	59.3±10.7	23.0±3.2	-0.1 ±3.8	0.89**	NR	0.78-0.81	NR	NR

*P<.05

**P<.01

c. 95% LoA: 95% limits of agreement

1. Body Density was measured instead of %BF

Potential errors for differences between the Bod Pod and DXA

Error in body surface area measurement

There can be measurement errors attributed to improper attire (S. Ball & Altena, 2004). As suggested by the manufacturer (Dempster & Aitkens, 1995), compressing clothes such as bathing suits and swimming caps are preferred to maintain the adiabatic conditions. Any clothes, hair, jackets, etc. show an apparent negative volume when measured in the chamber (Dempster & Aitkens, 1995). Therefore, any inappropriate attire can result in inaccuracy of the measurement. Under this situation, there may be overestimation of body density, which in turn results in underestimation of %BF (Dempster & Aitkens, 1995). Higgins et al. indicated that the presence of scalp and facial hair was shown to cause an underestimation of body fat by approximately 3% with approximately 1% due to facial hair and 2.3% due to scalp hair (Higgins, Fields, Hunter, & Gower, 2001). However, in the study that reported underestimated %BF, all participants had appropriate attire as recommended by the Bod Pod manufacturer (a swimsuit and a swim cap) in an effort to avoid such errors (Sardinha, et al., 1998). Therefore, the error might be attributed to reasons stated below.

Error in V_{TG} measurement/prediction

Another source of potential error might be the thoracic gas volume measurement/prediction (V_{TG}) (Sardinha, et al., 1998; Wagner & Heyward, 1999). V_{TG} involves the volume of air in the lungs and any air trapped in the thorax (Wagner & Heyward, 1999). The manufacturer assumed that body volume needs to be increased by

40% of V_{TG} in order to account for the difference between isothermal air in the thorax and adiabatic air in the Bod Pod chamber (Dempster & Aitkens, 1995). V_{TG} can either be measured or predicted. V_{TG} measurement is performed after body volume measurement when the subject takes a few normal tidal breaths followed by puffing against the occluded tube. Wagner et al. suggested that errors may exist in V_{TG} measurement since subjects are signaled to puff maneuver and occlude the airway during mid-exhalation rather than at the end of an exhalation. This results in an overestimation of V_{TG}. This in turn leads to overestimation of %BF (Wagner & Heyward, 1999).

Bod Pod also allows for the prediction of V_{TG}. The equation used to predict lung volume is based on functional residual capacity (FRC) predictions from the heights and ages of subjects aged 17-91 yrs (Crapo et al. 1982). Some studies compared predicted V_{TG} with measured V_{TG} in different populations including children, adults and athletes (Collins et al., 1999; Lockner, Heyward, Baumgartner, & Jenkins, 2000; McCrory, Mole, Gomez, Dewey, & Bernauer, 1998). McCrory et al. reported no significant difference between predicted and measured V_{TG}, while other researchers (Collins, et al., 1999; Lockner, et al., 2000{Collins, 1999 #45) showed large variation in the results (344-400ml), which implied that the prediction equation would not be applicable to every population (i.e. older adults).

On the other hand, Ball and colleagues proposed that predicted V_{TG} should not have a large influence on the final measured %BF (S. Ball & Altena, 2004). This is because according to the equation used to estimate Body Volume, (Body Volume = Body Volume_{raw} – Surface Area Artifact + $40\%V_{TG}$), only 40% of the V_{TG} is added to the body volume (S. Ball & Altena, 2004). Therefore, despite error in V_{TG} measurement/prediction, it should not have a large influence on the accuracy of total body volume measurement.

Error related to the two compartment model

The most highly debated assumption for ADP has been whether or not the FFM component is of constant density. Several researchers have found that the density of FFM varies based on exercise training, race, age and gender (Bunt, Going, Lohman, Heinrich, & Perry, 1990; Cote & Adams, 1993; Schutte et al., 1984). The 2 compartment (2C) model divides body composition into two compartments: fat mass (FM) and FFM, and assumes that their densities are relatively constant, namely 0.9g/cc for FM, and 1.10g/cc for FFM (Chumlea & Baumgartner, 1989). However, race, age, and inter-individual variability in FFM mineralization might affect the validity of this model. For example, Bentzur et al. (Bentzur, 2008) found that Bod Pod overestimated %BF in lean participants (BMI 21.65±2.0) when compared with DXA, while Ball et al. (S. Ball & Altena, 2004) found that the amount of difference in %BF between DXA and Bod Pod increased as body fat increased ($p < 0.0001$). Compared with the 2C model, a multi-component model such as the four-component model (4C), subdivides the FFM into four parts, namely water, protein, mineral, and fat tissue. It has been considered to be the most accurate of all body composition techniques (Ellis, 2000).

The 2C model is especially debatable when it is applied to older adults since body composition changes along with aging (Baumgartner, 1991) (i.e., hydration status, muscle mass and BMC loss (Wellens et al., 1994)). These changes may induce errors in %BF estimation. The two studies (Bosy-Westphal, 2003; Koda, et al., 2000) that were conducted with older adults suggested an association between age and measured %BF. Bosy-Westphal et al. (Bosy-Westphal, 2003) noted that the bias in %BF measurement by Bod Pod is mainly related to the water content of FFM. They indicated that a correction factor for total body water may improve the accuracy of ADP measurements in elderly females. This was supported by the evidence that a %BF difference between Bod Pod and DXA became larger when participant's body percent fat increased. While in Koda's study, the results suggested that the difference in BF% is positively correlated with age and negatively associated with BMC/FFM (Koda, et al., 2000), which might be explained by the increase in the hydration of FFM throughout normal aging (Schoeller, 1989).

Principles of bioelectrical impedance analysis

While laboratory methods (Hydrodensitometry, ADP, DXA) are generally more precise, they are also more time-consuming, inconvenient, and costly than field methods such as bioelectrical impedance analysis (BIA) (Wagner & Heyward, 1999). BIA is a relatively simple method for body composition assessment. It is quick, does not require a high degree of technician skill, and does not intrude on the client's privacy—even less invasive than skinfolds. Therefore, for older and obese individuals that have loose connective tissue or large fat-folds, it is the preferred field method (Wagner & Heyward, 1999).

The underlying principle of BIA is to determine body composition by introducing a flow of electrical current into the human body. Fat free mass, which contains large amounts of water and electrolytes, is a good electrical conductor; while fat, which is anhydrous, is an insulator (Kushner, 1992). FFM can be determined by measuring the resistance of a certain body region/whole body, which is proportional to the amount of water bounded to fat-free mass (typically 73% under normal hydration status) (Heyward, 1998). Therefore, fat mass can be estimated by subtracting FFM from the total body weight (Heymsfield, Wang, Visser, Gallagher, & Pierson, 1996).

Different BIA devices have been applied to clinical use including single frequency BIA and multi-frequency BIA. Single frequency devices typically introduce a flow of a low-level electrical current (800 mA) into the human body at a fixed frequency (50kHz; National Institutes of Health, 1994), while multi-frequency devices use different frequencies (0-500 kHz) to evaluate different parts of body fluid (Kyle, et al., 2004). However, whether one device is superior to the other is not yet determined.

Validity of bioelectrical impedance analysis

Although BIA has been widely used in body composition measurement in various populations, attention needs to be paid to the following limitations.

Assumptions of BIA

BIA is based on the assumptions that the human body is an isotropic conductor with a uniform length and cross-sectional area, and that current density is assumed to be uniformly distributed along axes in all directions, which is not true with humans (Kushner, 1992). These assumptions usually apply to single frequency BIA devices that measure whole body impedance. It has been claimed that multi-frequency and segmental BIA devices may yield better results in body composition measurement because 1) multi-frequency device introduces currents of different frequencies to the human body which differentiates intracellular and extracellular fluid; 2) segmental BIA device measures body composition in different segments (usually distal and trunk), which might be more accurate instead of measuring whole body impedance (Kyle, et al., 2004). However, no agreement was reached on the superiority of different devices (Kyle, et al., 2004).

Hydration status

Since BIA it is based on impedance to electrical current flow of the human body, the greater the total body water and FFM, the less resistance to the flow of the electric current (Wagner & Heyward, 1999). Therefore, participant’s hydration status can influence the accuracy of the measurement.

Several factors could contribute to hydration status, including age, obesity, and training level. For example, BIA significantly overestimated %BF by 6.4 ± 0.5% in a groups of endurance and power athletes ($p < .001$) because athletes have a higher proportion of lean body mass and therefore more water content (Company et al. 2010). Similarly, power lifters are reported to have lower impedance values when compared with people who are not involved in regular strength training (Huygens et al., 2002). On the other side, for obese individuals (BMI>30), BIA may underestimate %BF by approximately 3% (Shafer, 2009; G. Sun, 2005), and that the tendency of underestimation in FM tends to reduce along with weight loss (Savastano, 2010). For older adults, changes in body composition happen with aging, including a reduction in lean body mass, modifications in the amount of minerals in lean body mass, and intra-/extracellular water ratio, which could affect the outcomes (Fontana & Klein, 2007;

Forbes, 1999). Ravaglia et al. suggested that BIA overestimated %BF in subjects aged from 20-95, while the bias increased with age starting from 50 yrs, which might be attributed to increased hydration of fat free mass and variability in BMC throughout normal aging (Ravaglia, et al., 1999).

Choice of equations (aging, race)

Since BIA equations are developed from different populations (e.g., different ethnicities, ages, training levels and health conditions), choosing an appropriate BIA equation for the target population is very important. However, this at the same time can be a limitation factor for BIA (Kyle, et al., 2004). Some commonly used equations that are validated against reference standards (i.e. multi-compartment model) include the following formulas.

Deurenberg: $FFM\ (kg) = 0.671\ ht^2/R + 3.1\ sex + 3.9$ *(Deurenberg, Van der Kooij, Evers, & Hulshof, 1990)*

Baumgartner: $FFM\ (kg) = -1.73 + 0.28\ ht^2/R + 0.27\ wt + 4.5\ sex + 0.31\ leg\ circumference$ *in cm (Baumgartner, 1991)*

Roubenoff: $FFM\ (kg) = 7.74 + 0.45\ ht^2/R + 0.12\ wt + 0.05\ reactance$ *(Women)*

$FFM\ (kg) = 9.15 + 0.43\ ht^2/R + 0.20\ wt + 0.07\ reactance$ *(Men) (Roubenoff et al., 1997)*

Kyle: $FFM\ (kg) = -4.104 + 0.518\ height^2/R + 0.231\ wt + 0.130\ reactance + 4.229\ sex$ *(Kyle, Genton, Karsegard, Slosman, & Pichard, 2001)*

NHANES:

$FFM\ (kg) = -10.678 + 0.262\ weight + 0.652\ height^2/R + 0.015\ R$ *(Men)*

$FFM\ (kg) = -9.529 + 0.168\ weight + 0.696\ height^2/R + 0.016\ R$ *(Women) (Chumlea et al., 2002)*

where ht = height in cm, wt = weight in kg, R = resistance and sex is coded 0 for women and 1 for men.

Genton et al. (Genton, et al., 2001) suggested that Kyle's formula showed the best agreement with DXA when estimating FFM (limits of agreement –3.25-3.25 kg, SEE=1.6, r=0.89 for women; limits of agreement –3.77-4.15 kg, SEE=2.0, r=0.94 for men). The Deurenberg and Roubenoff formulas underestimated FFM when compared to DXA in both men and women, while Baumgartner formula overestimated fat free mass in both genders (Genton, et al., 2001). The NHANES III equation also has good precision. It is validated against the 4 compartment model when based on a large sample size (1474 whites and 355 blacks, aged 12-94 yrs) (S. S. Sun, 2003).

Conclusion

The review of literature suggests that since body composition can only be estimated rather than measured, each estimation tool has its limitations and assumptions that can not be applied to all ethnic or age groups, even with the use of technologically sophisticated methods (i.e., Bod Pod, DXA). While there are a variety of measurement methods, choosing an appropriate method for the target population helps increase the accuracy. BIA and ADP are convenient measurements of body composition that are not invasive for older adults. While the two measurements are validated against reference methods in younger individuals as well as children, whether they serve as valid measurements for older adults has not been concluded. Therefore, follow-up studies are necessary to further validate the two measurements.

CHAPTER 3 METHODS

Participants

A total of 64 older adults (40 females and 24 males) between 60 and 96 yrs were recruited from 2 mid-west communities (South Dakota and Iowa). Participants consist of 61 Caucasians and 3 Asians recruited for an exercise and nutrition intervention study designed to counteract muscle mass loss. Their medical history was obtained prior to the start of the study. Exclusion criteria were participants 1) with a history of liver or kidney diseases, morbid obesity, or endocrine disease and 2) with osteoporosis and/or chronic diseases that affect calcium or bone metabolism. The study was approved by the South Dakota State University Institutional Review Board for Human Subjects and Iowa State University Institutional Review Board for Human Subjects.

Measurements

Dual Energy X-ray Absorptiometry (DXA)

DXA (Hologic Discovery v.12.3) was used to measure body composition including fat mass (FM), fat free mass (FFM), and bone mineral densities (BMC). The device was calibrated using a phantom spine containing composites of bone, fat and lean tissue before participants were tested. Participants lay supine with arms and legs at their sides during the scan. One trained operator was responsible for conducting and analyzing the scans for all the participants. Manufacturer’s software version V12.3 was used for analysis of FFM and %BF.

Bioelectrical Impedance Analysis (BIA)

FFM and %BF were also determined by BIA. All subjects were fasted for 12 hours before the test. The measurement of BIA (Body Composition Analyzer, BIA-101S, RJL Systems, Clinton Township, MI) was carried out with the participants lying in a supine position on a flat, non-conductive bed. Two electrodes were placed both on the right wrist and ankle. A flow of a low-level electrical current (800 mA) was delivered into the participant's body at a fixed frequency (50 kHz). Kyle's equation (Kyle, et al., 2001) was used to calculate FFM:

$$FFM = -4.104 + (0.518 * Height^2/Resistance) + (0.231 * weight) + (0.130 * Reactance) + (4.229 * sex;\ Male = 1, Female = 0)$$

Air Displacement Plethysmography (Bod Pod)

Air displacement plethysmography was performed using the Bod Pod (BOD POD®, LMI, Concord CA). Prior to the testing, Bod Pod was calibrated using the following procedures: 1) calibrating volume of the empty chamber to establish the baseline and 2) calibrating the volume of a calibration cylinder of approximately 50L to establish range. Upon entering the chamber, all participants wore a tight-fitting swimsuit and a swim cap. Participants were instructed to sit motionless during body volume measurement. Body volume was measured twice. Thoracic gas volume (V_{TG}) was predicted using the age-specific prediction equation developed by Crapo et al. (Crapo, Morris, Clayton, & Nixon, 1982). Body density was calculated from the equations from the manufacturer

(Dempster & Aitkens, 1995). Percent body fat (%BF) was calculated from body density using the Siri equation (Siri, 1956):

$Body\ Surface\ Area\ (cm^2) = 71.84 * Weight\ (kg)^{0.425} * Height\ (cm)^{0.725}$

$Surface\ Area\ Artifact = k(l/cm^2) * Body\ Surface\ Area\ (cm^2)$, *where k is a negative constant derived by the manufacturer.*

$V_{TG}\ (L) = Functional\ Residual\ Capacity + 0.5Tidal\ Volume$

$Functional\ Residual\ Capacity\ (L) = 0.0472\ (height\ in\ cm) + 0.0090\ (age) – 5.290\ (women)$

$Functional\ Residual\ Capacity\ (L) = 0.0360\ (height\ in\ cm) + 0.0031\ (age) – 3.182\ (men)$

$Body\ Volume\ (L) = Body\ Volume_{raw} – Surface\ Area\ Artifact + 40\%V_{TG}$

$Body\ Density\ (kg/L) = Body\ Weight/Body\ Volume$

$\%BF = (495/body\ density) – 450$ *(Siri, 1956)*

Statistical analysis

All statistical analyses employed herein were performed using SPSS 17.0. Data were examined for normal distributions using the Kolmogorov-Smirnov test. The comparisons of Bod Pod and BIA was assessed using paired sample t-test, Pearson correlation coefficients, simple linear regression, Cohen’s d, and absolute percent errors (i.e., APE_{BIA}=|BIA-DXA|/DXA*100). Two by three two way analyses of variance () were used to detect gender differences and interactions between gender and measurement methods. Bland-Altman plots were used to investigate systematic/proportional biases in comparisons between different methods (i.e. Bod Pod vs. DXA and BIA vs. DXA) for body composition measurement. Sensitivity and specificity as well as Kappa statistics was used to determine the agreement of BIA and ADP relative to DXA in classifying obesity (males and females over 55yrs with %BF >23% or >35% are considered obese) (Heyward, 2006). All tests were two-tailed and significance level was set at 0.05.

CHAPTER 4 RESULTS

Physical characteristics of the participants

Descriptive characteristics of the participants (40 males, 24 females) are listed in Table 1. Participants (61 Caucasians, 3 Asian) are aged from 60 to 96yrs, with BMI ranging from 17.6 to 54.3. There are significant sex differences for all descriptive characteristics of the participants except for age, BMI and reactance.

Comparison of %BF measured by BIA and ADP against DXA

The comparisons of BIA and ADP against DXA are summarized in Table 2. Group mean overestimation of %BF by ADP and BIA in relation to DXA was observed for both genders. However, ADP had smaller difference with DXA than BIA, supported by relatively smaller absolute percent errors (14.1±12.6% for BIA, 12.4±8.6% for ADP) as well as smaller Cohen's d (0.40 for BIA, 0.35 for ADP). The correlation coefficient between ADP and DXA was higher ($r = 0.95$, $p<0.05$) than BIA ($r = 0.85$, $p<0.05$) in both genders (Table 2). Both ADP (Slope = 1.24, SEE=3.23, $R^2=0.89$) and BIA (Slope = 1.02, SEE=4.78, $R^2 = 0.72$) showed good linear relationship with DXA; however, ADP had a smaller SEE indicating a smaller variance in the measurement (Figure 1). According to Bland-Altman plot A in Figure 2, no specific systematic bias ($p=0.83$) was observed for the comparison between BIA and DXA, with limits of agreement ranging from -6.0% to 12.6%. However, according to Bland-Altman plot B in Figure 2, a positive linear trend (Slope= 0.24, $R^2=0.24$, $p<0.05$) was detected, indicating a systematic pattern of %BF overestimation when participant's adiposity (%BF measured by DXA) was taken into consideration (limits of agreement ranged from -4.1% to 10.3%).

The difference in %BF between BIA and DXA was greater in females than in males, supported by a larger Cohen's d (0.58 for females, 0.40 for males). Similar gender differences were detected in ADP, where the overestimation of %BF was more significant in females than males (Cohen's d = 0.58 for females, 0.27 for males). However, independent sample t-test showed that the overestimation in females is not different from males in both ADP (*p*=0.06) and in BIA (*p*=0.30). Two way analysis of variance (2x3 ANOVA with gender and method as the independent variable) showed no significant interaction between gender and methods, which means the magnitude of gender differences do not interact with difference measurement methods used.

Classification agreement in obesity among BIA, ADP and DXA

Values for obesity classification agreement among the three measurements are listed in Table 2. The sensitivity and specificity of BIA in relation to DXA were 82.7% and 58.3%, respectively. Males (89.7%) had higher sensitivity than females (78.8%). ADP showed relatively higher sensitivity (86.8%) and specificity (81.8%) than BIA, with males (90.0%) higher than females (84.6%). Kappa statistics showed fair agreement between BIA and DXA (Kappa=0.36, 95%CI [0.10, 0.63]), and that males showed higher agreement (Kappa=0.50, 95%CI [0.07, 0.92]) than females (Kappa=0.29, 95%CI [-0.04, 0.62]). ADP indicated moderate agreement (Kappa=0.58 95%CI [0.34, 0.82]) with DXA, which is higher than BIA, with no gender difference.

Results: Tables and Figures

Table 1. Descriptive characteristics of the participants (N=62)

	All	Male	Female	p-value
Descriptive				
Age (y)	71±9	73±9	70±9	0.23
Height (cm)	161±15.1	174.3±10.4	152.6±11.2	<.01
Weight (kg)	78.0±19.8	90.9±22.8	70.2±12.9	<.01
BMI (kg/m^2)	30.2±6.3	30.0±7.2	30.4±5.7	0.81
Resistance	533.1	469.6±35.7	571.3±62.5	<.01
Reactance	55.9	53.8±22.9	57.2±11.0	0.50
%BF (%)				
BIA	36.9±9.0	30.2±8.4	40.9±6.6	<.01
ADP	36.7±9.8	29.3±8.6	41.2±7.5	<.01
DXA	33.6±7.5	27.3±6.0	37.4±5.4	<.01
FFM (kg)				
BIA	48.9±12.5	62.0±8.8	41.0±6.3	<.01
ADP	45.3±14.2	57.6±12.3	37.9±9.3	<.01
DXA	51.5±11.6	63.7±7.8	44.1±5.6	<.01

All values are Mean±SD; BMI, body mass index; BIA, bioelectrical impedance analysis; ADP, air-displacement plethysmography; DXA dual energy X-ray absorptiometry; %BF, percent body fat; FFM, fat-free mass.

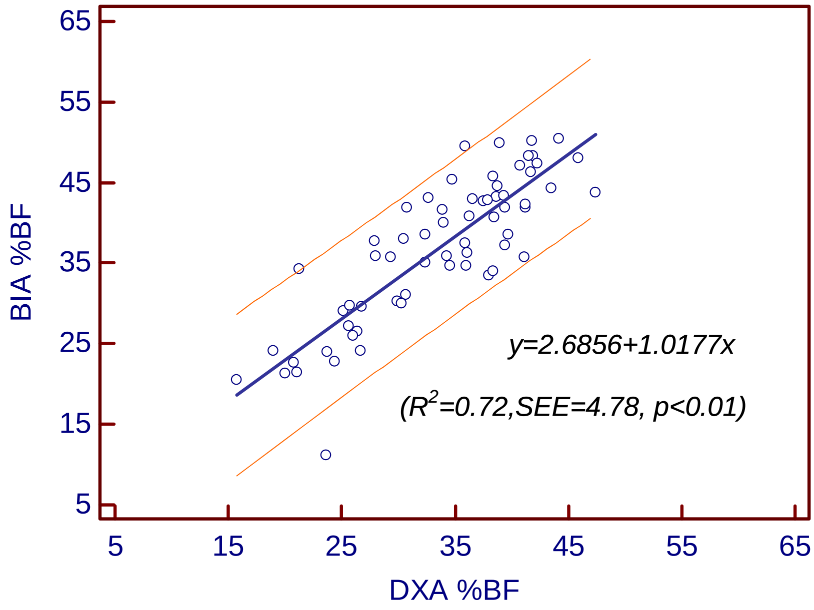
Table 2. Comparison of body percent fat measured by bioelectrical impedance analysis (BIA) and air-displacement plethysmography (ADP) against dual energy X-ray absorptiometry (DXA)

	Mean difference	Absolute percent error%	Cohen's d	r	Slope	R^2	SEE	Obesity classification accuracy		
								Sensitivity	Specificity	Kappa (95%CI)
BIA vs. DXA										
Female	3.5 ± 4.4*	12.7± 9.1	0.58	0.75*	0.92*	0.56	4.45	78.8%	57.1%	0.29 (-0.04, 0.62)
Male	2.9 ± 5.3*	16.5 ± 16.9	0.40	0.77*	1.08*	0.60	5.42	89.7%	60.0%	0.50 (0.07, 0.92)
All	3.3 ± 4.7*	14.1±12.6	0.40	0.85*	1.02*	0.72	4.78	82.7%	58.3%	0.36 (0.10, 0.63)
Bod Pod vs. DXA										
Female	3.8 ± 3.5*	11.7 ± 7.9	0.58	0.91*	1.27*	0.83	3.19	84.6%	85.7%	0.58 (0.28, 0.87)
Male	2.0 ± 3.8*	13.5 ± 9.8	0.27	0.92*	1.32*	0.85	3.37	90.0%	75.0%	0.59 (0.18, 1.00)
All	3.1 ± 3.7*	12.4 ± 8.6	0.35	0.95*	1.04*	0.89	3.23	86.8%	81.8%	0.58 (0.34, 0.82)

Absolute percent error% = APE=|BIA-DXA|/DXA*100; Cohen’s d, magnitude of differences between ADP, BIA against DXA; r, correlation coefficient; SEE, standard error of estimation; Sensitivity, proportion of participants that are obese according to BIA and ADP that are correctly identified as such with DXA; Specificity, proportion of participants that are non-obese according to BIA and ADP that are correctly identified as such with DXA. * P<.01

Figure 1. Scatter plot for the linear relationship between %BF measured by BIA, ADP and DXA

A. BIA vs. DXA



B. ADP vs. DXA

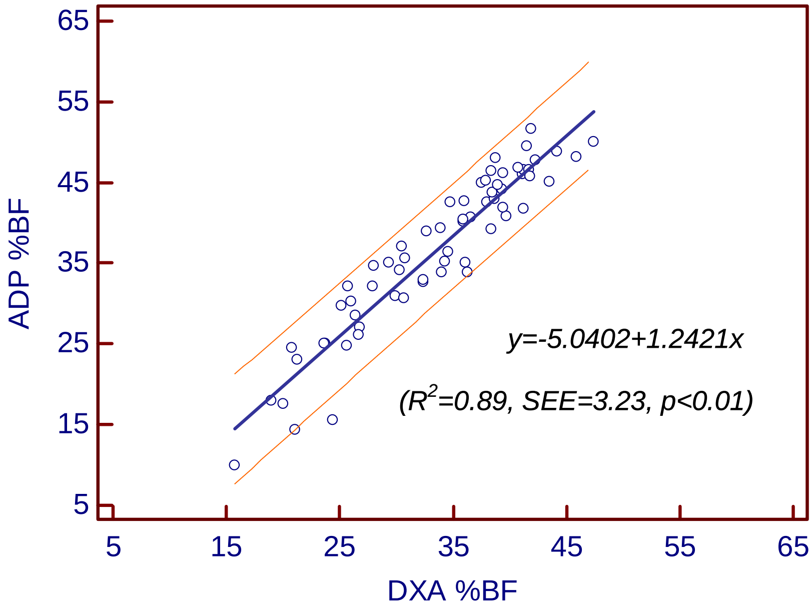
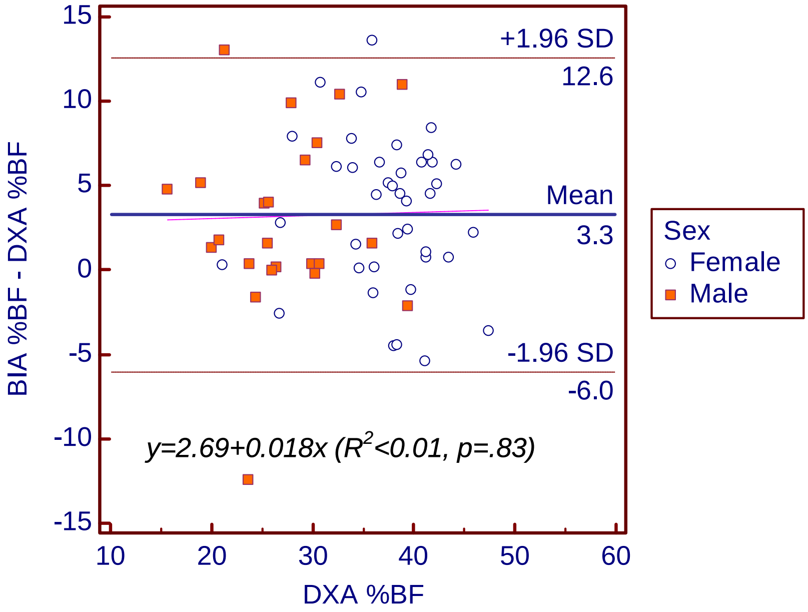
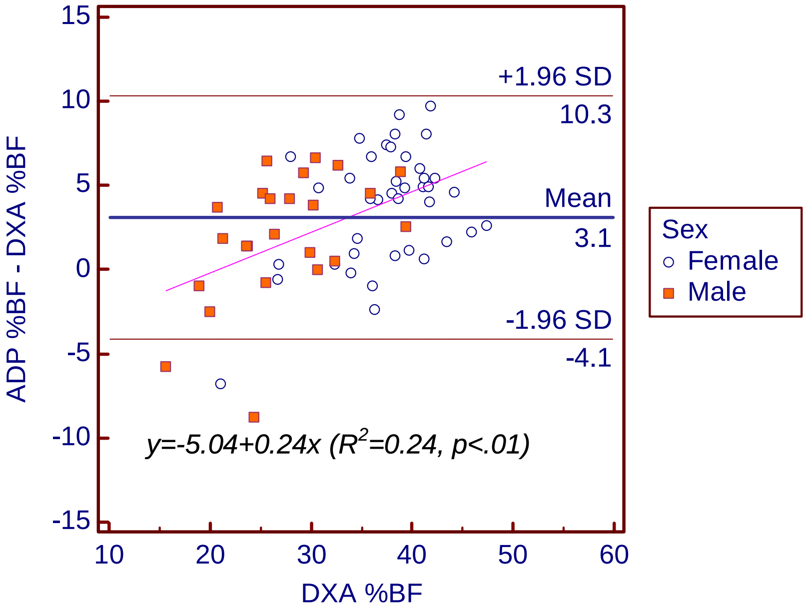


Figure 2. Bland-Altman plot for the relationship between %BF measured by BIA, ADP and DXA

A. BIA vs. DXA



B. ADP vs. DXA



CHAPTER 5 DISCUSSION

Loss of muscle strength and mass that happens along with aging is becoming an issue of great interest in older adults. For nutrition/exercise interventions that may help counteract this process, an accurate measurement of body composition is essential to evaluate the effectiveness of the interventions. However, although DXA and HW have long been considered the ‘gold standards’, some drawbacks have limited their widespread use in research (e.g., costly to perform, inconvenient for older adults who may have difficulties submerging under water). In the meantime, BIA and ADP are non-invasive measurements that are easy to perform for this population. In this study, the validity of BIA and ADP was evaluated against DXA.

The current study found that %BF was significantly overestimated by both BIA and ADP in comparison to DXA. However, ADP had greater agreement relative to DXA than BIA based on smaller Cohen’s d and absolute percent errors as well as SEE (indication of variations). Similar results were reported in previous studies. For example, Moon et al. suggested that BIA had larger variations (SEE=4.7) than ADP (SEE=3.3) in the estimation of %BF for high school boys when HW was used as the reference standard. Also, Sardinha et al. (Sardinha, et al., 1998) reported that the %BF prediction model that included ADP plus age, weight and height showed better predictive ability (R^2=0.895, SEE=2.4) than the model that used a combination of BIA plus age, weight and height (R^2=0.798, SEE=3.3). The current study also showed better classification agreement in separating obese from non-obese participants with ADP in comparison with BIA. Noted that although BMI has also been widely used to classify obesity (BMI>30 is classified as obese (World Health Organization, 2007)), it showed lower specificity (38.00%) and Kappa score (0.27) than BIA and ADP despite a similar sensitivity in the current study. ADP had a significantly proportional bias related to participants' body fatness. Similar bias was reported by Ball et al. (S. D. Ball, 2004) and Levenhagen et al. (Levenhagen, Borel, Welch, Piasecki, & Piasecki, 1999) that although ADP and reference standards (i.e. DXA, HW) were highly correlated, the amount of difference increased as participants' body fatness increased. Therefore, practitioners should be cautious when using ADP to estimate %BF in obese individuals.

The current study confirmed the results of some previous studies (Genton, et al., 2001; Ramel, Geirsdottir, Arnarson, & Thorsdottir, 2011; Ravaglia, et al., 1999) that reported overestimation of %BF with BIA. In contrast, some other studies (Braulio, 2010; Leahy, et al., 2012; Savastano, 2010; G. Sun, 2005) found that BIA underestimated %BF in relation to DXA. There are several possible explanations for the inconsistent outcomes of the current study with previous studies. First of all, different types of BIA devices were used. Previous studies that used other types of BIA showed different results from the current study in which a single frequency whole body BIA device was used. For example, Sun et al. (G. Sun, 2005) reported that multi-frequency BIA overestimated %BF for lean subjects (%BF<15% for men and <33% for women) while underestimated %BF for obese subjects (%>25 for men and >33% for women). Also, Leahy et al. (Leahy, et al., 2012) showed underestimated %BF when regional BIA was used. Secondly, different equations for the estimation of FFM were applied. Both Ramel et al. (Ramel, et al., 2011) and Genton et al. (Genton, et al., 2001) reported that the validity of BIA was different when using various equations. The equation used in this study was developed by Kyle et al (Kyle, et al., 2001). This equation was proved to be the most accurate for older adults among currently commonly used equations (e.g., Deurenberg equation, Baumgartner equation and Roubenoff equation) (Genton, et al., 2001). Another possible reason is that there are variations in the populations involved. Studies that found underestimation of %BF (Braulio, 2010; Leahy, et al., 2012; Savastano, 2010; G. Sun, 2005) focused on adults or younger adults aged under 55yrs, while overestimation of %BF (Genton, et al., 2001; Ramel, et al., 2011; Ravaglia, et al., 1999) was observed in studies where participants aged over 55yrs were included. The current study was focused on older adults; therefore, the findings confirmed that BIA would likely to overestimate %BF in this population. BIA assumes a fixed percentage of water in FFM (Chumlea & Baumgartner, 1989).

With respect to the comparison between ADP and DXA, ADP significantly overestimated %BF. This also confirmed the findings of previous studies (S. D. Ball, 2004; Bosy-Westphal, 2003; Wagner, et al., 2000). The mean difference (3.1±3.7%) in the current study, however, was greater than that (i.e., 1.8%) of the previous studies. One possible explanation for this inconsistency is the errors related to the predicted $V_{TG.}$ This study used the equation provided by the manufacturer to predict V_{TG} which was developed based on functional residual capacity predictions by Crapo et al. from the heights and ages of 245 healthy nonsmoking subjects aged 17-91 yrs (Crapo, et al., 1982). The equation may not be generalizable for older adults since it is not developed specifically for this population. It was reported that the error of predicted V_{TG} can be up to 400ml (Collins et al. 1999, Lockner et al. 2000), which means for a 70kg person with a body density of 1.04kg/cc, an error of 400ml in

V_{TG} can result in an absolute %BF error of about 4.6%. Another possible reason might be attributable to the use of the 2C model, which may elicit more incorrect values of body composition when applied to older adults than younger adults (Heyward, 2006). Based on a two-compartment model, ADP assumes that the human body consists of FM and FFM, and that each part has a fixed density (Chumlea & Baumgartner, 1989). However, the density of FM or FFM varies in different populations (i.e. the average density of the FFM of older adults is 1.098g/cc, which is lower than the assumed 1.100g/cc by the 2C model). Thus, the %BF of older adults may be systematically overestimated (Heyward, 2006).

In the current study, males showed smaller differences as well as better classification agreements in relation to DXA than females in both BIA and ADP. This was also observed in the study of Koda et al. (Koda, et al., 2000) that the difference between ADP and DXA was larger in females than in males when ADP was validated against DXA. A possible explanation for this gender difference is that errors are more likely to occur when participants are more obese; females had significantly higher %BF than males in the current study (Levenhagen, et al., 1999).

Some limitations of this study need to be addressed. First of all, since all participants were recruited for a nutrition and exercise intervention study to counteract muscle strength/mass loss, they all had a serum 25OH-VitD$_3$ concentration of 10-25ng/ml, which is a low vitamin D level. Therefore, it would be better if the population were more diverse. Moreover, the use of predicted rather than measured VTG might results in errors related to %BF measurement using ADP. In addition, although all the participants wore a tight fitting suit as well as a swimming cap as required by the manufacturer during measurement

(Dempster & Aitkens, 1995), facial hair could not be controlled for some male participants. However, it was reported in a previous study (Higgins, et al., 2001) that this could cause only 1% underestimation of %BF (Higgins, et al., 2001).

In conclusion, the current study plays a key role in creating consensus on which method – either BIA or ADP – would more accurately measure %BF and classify obesity in older populations. Given that both BIA and ADP had acceptable agreement (with overestimation of about 3.2%BF) relative to the reference standard (i.e. DXA) in older adults, they all appear to be able to serve as alternatives to DXA in measuring %BF. In addition, ADP showed slightly better agreement in relation to DXA in comparison to BIA in %BF measurement and obesity classification. However, when using ADP to estimate body composition in obese older adults, researchers and/or practitioners should be cautious of the potential systematic bias (as demonstrated with Bland-Altman plots). Future research is needed to test the validity of ADP and BIA in healthy older adults with diverse ethnicity and adiposity.

REFERENCES

Aleman-Mateo, H., Huerta, R.H., Esparza-Romero, J, Mendez, R.O., Urquidez, R., & Valencia, M.E. (2007). Body Composition by the Four-Compartment Model: Validity of the Bod Pod for Assessing Body Fat in Mexican Elderly. *European Journal of Clinical Nutrition, 61*(7), 830-836.

Arngrimsson, S. A., Evans, E. M., Saunders, M. J., Ogburn, C. L., Lewis, R. D. (2000). Validation of Body Composition Estimates in Male and Female Distance Runners Using Estimates from a Four-Component Model. *American Journal of Human Biology, 12*(3), 301-314.

Ball, S. D., Altena, T. S. (2004). Comparison of the Bod Pod and Dual Energy X-Ray Absorptiometry in Men. *Physiological Measurement, 25*(3), 671-678.

Ballard, T. P., Fafara, L., & Vukovich, M. D. (2004). Comparison of Bod Pod (R) and Dxa in Female Collegiate Athletes. *Medicine and Science in Sports and Exercise, 36*(4), 731-735.

Baumgartner, R. N. (1991). Body Composition in Elderly People: Effect of Criterion Estimates on Predictive Equations. *The American journal of clinical nutrition, 53*(6), 1345-1353.

Baumgartner, R. N. (2000). Body Composition in Healthy Aging. *Annals of the New York Academy of Sciences, 904*(1), 437-448.

Benton, M. J. (2011). Sarcopenic Obesity: Strategies for Management. *American journal of nursing, 111*(12), 38-44.

Bentzur, K. M. (2008). Evaluation of the Bod Pod for Estimating Percent Body Fat in Collegiate Track and Field Female Athletes: A Comparison of Four Methods. *Journal of Strength and Conditioning Research, 22*(6), 1985-1991.

Bosy-Westphal, A. (2003). Validation of Air-Displacement Plethysmography for Estimation of Body Fat Mass in Healthy Elderly Subjects. *European Journal of Nutrition, 42*(4), 207-216.

Braulio, V. B. (2010). Comparison of Body Composition Methods in Overweight and Obese Brazilian Women. *Arquivos Brasileiros de Endocrinologia e Metabologia, 54*(4), 398-405.

Bunt, J. C., Going, S. B., Lohman, T. G., Heinrich, C. H., & Perry, C. D. (1990). Variation in Bone-Mineral Content and Estimated Body-Fat in Young-Adult Females. *Medicine and science in sports and exercise, 22*(5), 564-569.

Chumlea, W. C., & Baumgartner, R. N. (1989). Status of Anthropometry and Body Composition Data in Elderly Subjects. *The American Journal of Clinical Nutrition, 50*(5), 1158-1166.

Chumlea, W. C., Guo, S. S., Kuczmarski, R. K., Flegal, K. M., L., Johnson C., B., Heymsfield S., Lukaski, H. C., Friedl, K., Hubbard V. S. (2002). Body Composition Estimates from Nhanes Iii Bioelectrical Impedance Data. *International Journal of Obesity, 26*(12), 1596-1609.

Collins, M. A., Millard-Stafford, M. L., Sparling, P. B., Snow, T. K., Rosskopf, L. B., Webb, S. A., & Omer, J. . (1999). Evaluation of the Bod Pod (R) for Assessing Body Fat in Collegiate Football Players. *Medicine and Science in Sports and Exercise, 31*(9), 1350-1356.

Company, Joe, & Stephen, Ball. (2010). Body Composition Comparison: Bioelectric Impedance Analysis with Dual-Energy X-Ray Absorptiometry in Adult Athletes. *Measurement in Physical Education and Exercise Science, 14*(3), 186-201.

Cote, K. D., & Adams, W. C. (1993). Effect of Bone-Density on Body-Composition Estimates in Young-Adult Black-and-White Women. *Medicine and Science in Sports and Exercise, 25*(2), 290-296.

Crapo, R. O., Morris, A. H., Clayton, P. D., & Nixon, C. R. (1982). Lung Volumes in Healthy Nonsmoking Adults. *Bulletin Européen de Physiopathologie Respiratoire, 18*(3), 419-425.

Dempster, P., & Aitkens, S. . (1995). A New Air Displacement Method for the Determination of Human Body Composition. *Medicine and Science in Sports and Exercise, 27*(12), 1692-1697.

Deurenberg, P., Van der Kooij, K. , Evers, P., & Hulshof, T. (1990). Assessment of Body Composition by Bioelectrical Impedance in a Population Aged Greater Than 60 Y. *The American Journal of Clinical Nutrition, 51*(1), 3-6.

Ellis, K. J. (2000). Human Body Composition: In Vivo Methods. *Physiological Reviews, 80*(2), 649-680.

Fields, D. A., Goran, M. I., & McCrory, M. A. (2002). Body-Composition Assessment Via Air-Displacement Plethysmography in Adults and Children: A Review. *The American Journal of Clinical Nutrition, 75*(3), 453-467.

Fontana, Luigi, & Klein, Samuel. (2007). Aging, Adiposity, and Calorie Restriction. *JAMA (Chicago, Ill.), 297*(9), 986-994.

Forbes, G. B. (1999). Longitudinal Changes in Adult Fat-Free Mass: Influence of Body Weight. *The American Journal of Clinical Nutrition, 70*(6), 1025-1031.

Genton, L., L., Karsegard V., Kyle, U. G. , Hans, D. B., Michel, J-P., & Pichard, C. (2001). Comparison of Four Bioelectrical Impedance Analysis Formulas in Healthy Elderly Subjects. *Gerontology, 47*(6), 315-323.

Ginde, S. R. (2005). Air Displacement Plethysmography: Validation in Overweight and Obese Subjects. *Obesity Research, 13*(7), 1232-1237.

Han, T. S., Carter, R., Currall, J. E., & Lean, M. E. (1996). The Influence of Fat Free Mass on Prediction of Densitometric Body Composition by Bioelectrical Impedance Analysis and by Anthropometry. *European Journal of Clinical Nutrition, 50*(8), 542-548.

Heymsfield, S. B., Wang, Z. M., Visser, M., Gallagher, D., & Pierson, R. N. (1996). Techniques Used in the Measurement of Body Composition: An Overview with Emphasis on Bioelectrical Impedance Analysis. *The American Journal of Clinical Nutrition, 64*(3), 478S-484S.

Heyward, V. H. (1998). Practical Body Composition Assessment for Children, Adults, and Older Adults. *International Journal of Sport Nutrition and Exercise Metabolism, 8*(3), 285-307.

Heyward, V. H. (2006). Advanced Fitness Assessment and Exercise Prescription.5th ed. Champaign (IL): *Human Kinetics*; 181-182.

Higgins, P. B., Fields, D. A., Hunter, G. R., & Gower, B. A. (2001). Effect of Scalp and Facial Hair on Air Displacement Plethysmography Estimates of Percentage of Body Fat. *Obesity Research, 9*(5), 326-330.

Himes, J. H. (1999). Agreement among Anthropometric Indicators Identifying the Fattest Adolescents. *Journal of Obesity, 23*(2), 18-21.

Holmes, Jason, Gibson, Ann, Cremades, J. G., & Mier, Constance. (2011). Body-Density Measurement in Children: The Bod Pod Versus Hydrodensitometry. *International Journal of Sport Nutrition and Exercise Metabolism, 21*(3), 240-247.

Huygens, W., Claessens, A. L., Thomis, M., Loos, R., & Van Langendonck, L. (2002). Body Composition Estimations by Bia Versus Anthropometric Equations in Body Builders and Other Power Athletes. *Journal of Sports Medicine and Physical Fitness, 42*(1), 45-55.

Jaffrin, M. Y., & Morel, H. (2008). Body Fluid Volumes Measurements by Impedance: A Review of Bioimpedance Spectroscopy (BIS) and Bioimpedance Analysis (BIA) Methods. *Medical Engineering & Physics, 30*(10), 1257-1269.

Kiebzak, G. M., Leamy, L. J., Pierson, L. M., Nord, R. H., & Zhang, Z. Y. (2000). Measurement Precision of Body Composition Variables Using the Lunar Dpx-L Densitometer. *Journal of Clinical Densitometry, 3*(1), 35-41.

Koda, M., Tsuzuku, S., Ando, F., Niino, N., & Shimokata, H. (2000). Body Composition by Air Displacement Plethysmography in Middle‐Aged and Elderly Japanese: Comparison with Dual‐Energy X‐Ray Absorptiometry. *Annals of the New York Academy of Sciences, 904*(1), 484-488.

Kushner, R. F. (1992). Bioelectrical Impedance Analysis: A Review of Principles and Applications. *Journal of the American College of Nutrition, 11*(2), 199-209.

Kyle, U. G., Bosaeus, I., De Lorenzo, A.D., Deurenberg, P., Elia, M., Gómez, J. M., . . . Pichard, C. (2004). Bioelectrical Impedance Analysis--Part I: Review of Principles and Methods. *Clinical Nutrition, 23*(5), 1226-1243.

Kyle, U. G., Genton, L., Karsegard, L., Slosman, D. O., & Pichard, C. (2001). Single Prediction Equation for Bioelectrical Impedance Analysis in Adults Aged 20–94 Years. *Nutrition, 17*(3), 248-253.

Laurson, K. R., Eisenmann, J. C., & Welk, G. J. (2011). Body Mass Index Standards Based on Agreement with Health-Related Body Fat. *American Journal of Preventive Medicine, 41*(4), S100-S105.

Leahy, Siobhan, O'Neill, Cian, Sohun, Rhoda, & Jakeman, Philip. (2012). A Comparison of Dual Energy X-Ray Absorptiometry and Bioelectrical Impedance Analysis to Measure Total and Segmental Body Composition in Healthy Young Adults. *European Journal of Applied Physiology, 112*(2), 589-595.

Levenhagen, D. K., Borel, M. J., Welch, D. C., Piasecki, J. H., & Piasecki, D. P. (1999). A Comparison of Air Displacement Plethysmography with Three Other Techniques to Determine Body Fat in Healthy Adults. *JPEN, Journal of Parenteral and Enteral Nutrition, 23*(5), 293-299.

Lockner, D. W., Heyward, V. H., Baumgartner, R. N., & Jenkins, K. A. (2000). Comparison of Air‐Displacement Plethysmography, Hydrodensitometry, and Dual X−Ray Absorptiometry for Assessing Body Composition of Children 10 to 18 Years of Age. *Annals of the New York Academy of Sciences, 904*(1), 72-78.

Mazess, R. B., Barden, H. S., Biesek, J. P., & Hanson, J. (1990). Dual-Energy X-Ray Absorptiometry for Total-Body and Regional Bone-Mineral and Soft-Tissue Composition. *The American Journal of Clinical Nutrition, 51*(6), 1106-1112.

McCrory, M. A., Mole, P. A., Gomez, T. D., Dewey, K. G., & Bernauer, E. M. (1998). Body Composition by Air-Displacement Plethysmography by Using Predicted and Measured Thoracic Gas Volumes. *Journal of Applied Physiology: Respiratory, Environmental and Exercise Physiology, 84*(4), 1475-1479.

Nissen, S., Sharp, R., Ray, M., Rathmacher, J. A., Rice, D. , Fuller, J. C., . . . Abumrad, N. (1996). Effect of Leucine Metabolite B-Hydroxy-B-Methylbutyrate on Muscle Metabolism During Resistance-Exercise Training. *Journal of Applied Physiology: Respiratory, Environmental and Exercise Physiology, 81*(5), 2095-2104.

Panotopoulos, G., Charles, R. J., Bernard, G-G., & Arnaud, B. (2001). Dual X-Ray Absorptiometry, Bioelectrical Impedance, and near Infrared Interactance in Obese Women. *Medicine and Science in Sports and Exercise, 33*(4), 665-670.

Panton, L. B., Rathmacher, J. A., Baier, S., & Nissen, S. (2000). Nutritional Supplementation of the Leucine Metabolite B-Hydroxy-B-Methylbutyrate (HMB) During Resistance Training. *Nutrition, 16*(9), 734-739.

Pietrobelli, A., Formica, C., Wang, Z. M., & Heymsfield, S. B. (1996). Dual-Energy X-Ray Absorptiometry Body Composition Model: Review of Physical Concepts. *American Journal of Physiology: Endocrinology and Metabolism, 271*(6), E941-E951.

Plank, L. D. (2005). Dual-Energy X-Ray Absorptiometry and Body Composition. *Current opinion in clinical nutrition & metabolic care, 8*(3), 305-309.

Prior, B. M., Cureton, K. J., Modlesky, C. M., Evans, E. M., & Sloniger, M. A. (1997). In Vivo Validation of Whole Body Composition Estimates from Dual-Energy X-Ray Absorptiometry. *Journal of applied physiology, 83*(2), 623-630.

Ramel, A., Geirsdottir, O. G., Arnarson, A., & Thorsdottir, I. (2011). Regional and Total Body Bioelectrical Impedance Analysis Compared with DXA in Icelandic Elderly. *European Journal of Clinical Nutrition, 65*(8), 978-983.

Ravaglia, G., Forti, P., Maioli, F., Boschi, F., Cicognani, A., & Gasbarrini, G. (1999). Measurement of Body Fat in Healthy Elderly Men: A Comparison of Methods. *The Journals of Gerontology. Series A, Biological Sciences and Medical Sciences, 54*(2), M70-M76.

Roubenoff, R., Baumgartner, R. N., Harris, T. B., Dallal, G. E., Hannan, M. T., Economos, C. D., . . . Kiel, D. P. (1997). Application of Bioelectrical Impedance Analysis to Elderly Populations. *The Journals of Gerontology. Series A, Biological Sciences and Medical Sciences, 52*(3), M129-M136.

Roubenoff, R., & Hughes, V. A. (2000). Sarcopenia Current Concepts. *The Journals of Gerontology. Series A, Biological Sciences and Medical Sciences, 55*(12), M716-M724.

Samper Ternent, Rafael, & Al Snih, Soham. (2012). Obesity in Older Adults: Epidemiology and Implications for Disability and Disease. *Reviews in Clinical Gerontology, 22*(1), 10-34.

Sardinha, L. B., Lohman, T. G., Teixeira, P. J., Guedes, D. P., & Going, S. B. (1998). Comparison of Air Displacement Plethysmography with Dual-Energy X-Ray Absorptiometry and 3 Field Methods for Estimating Body Composition in Middle-Aged Men. *The American Journal of Clinical Nutrition, 68*(4), 786-793.

Savastano, S. (2010). Validity of Bioelectrical Impedance Analysis to Estimate Body Composition Changes after Bariatric Surgery in Premenopausal Morbidly Women. *Obesity surgery, 20*(3), 332-339.

Schoeller, D. A. (1989). Changes in Total Body Water with Age. *The American Journal of Clinical Nutrition, 50*(5), 1176-1181.

Schutte, J. E., Townsend, E. J., Hugg, J., Shoup, R. F., Malina, R. M., & Blomqvist, C. G. (1984). Density of Lean Body Mass Is Greater in Blacks Than in Whites. *Journal of Applied Physiology: Respiratory, Environmental and Exercise Physiology, 56*(6), 1647-1649.

Shafer, K. J. (2009). Validity of Segmental Multiple-Frequency Bioelectrical Impedance Analysis to Estimate Body Composition of Adults across a Range of Body Mass Indexes. *Nutrition, 25*(1), 25-32.

Siri, W. E. (1993). Body Composition from Fluid Spaces and Density: Analysis of Methods. *Nutrition*, 9(5), 480-491

Sun, G. (2005). Comparison of Multifrequency Bioelectrical Impedance Analysis with Dual-Energy X-Ray Absorptiometry for Assessment of Percentage Body Fat in a Large, Healthy Population. *The American Journal of Clinical Nutrition, 81*(1), 74-78.
Sun, S. S. (2003). Development of Bioelectrical Impedance Analysis Prediction Equations for Body Composition with the Use of a Multicomponent Model for Use in Epidemiologic Surveys. *The American Journal of Clinical Nutrition, 77*(2), 331-340.

Thomson, J. S. (2009). Effects of Nine Weeks of [Beta]-Hydroxy-[Beta]-Methylbutyrate Supplementation on Strength and Body Composition in Resistance Trained Men. *Journal of Strength and Conditioning Research, 23*(3), 827-835.

Tseh, W., Caputo, J. L., & Keefer, D. J. (2010). Validity and Reliability of the Bod Pod® S/T Tracking System. *International Journal of Sports Medicine, 31*(10), 704-708.

Wagner, D. R., & Heyward, V. H. (1999). Techniques of Body Composition Assessment: A Review of Laboratory and Field Methods. *Research Quarterly for Exercise and Sport, 70*(2), 135-149.

Wagner, D. R., Heyward, V. H., & Gibson, A. L. (2000). Validation of Air Displacement Plethysmography for Assessing Body Composition. *Medicine and Science in Sports and Exercise, 32*(7), 1339-1344.

Waki, M. (1991). Relative Expansion of Extracellular Fluid in Obese Vs. Nonobese Women. *American journal of physiology: endocrinology and metabolism, 261*(2), E199-E203.

Wellens, R., Guo, S. M., Chumlea, W. C., Roche, A. F., Reo, N. V., & Siervogel, R. M. (1994). Body Composition in White Adults by Dual-Energy X-Ray Absorptiometry, Densitometry, and Total Body Water. *The American Journal of Clinical Nutrition, 59*(3), 547-555.

Yee, A. J., Fuerst, T., Salamone, L., Visser, M., Dockrell, M., Van Loan, M., & Kern, M. (2001). Calibration and Validation of an Air-Displacement Plethysmography Method for Estimating Percentage Body Fat in an Elderly Population: A Comparison among Compartmental Models. *The American Journal of Clinical Nutrition, 74*(5), 637-642.

APPENDIX—Raw Data

Raw Data—Descriptive

Subject ID	Sex	Age, (yrs)	Weight (kg)	Height (cm)	BMI	Resistance	Reactance
1019	0	70	73.0	168.0	25.9	588	71
1068	0	83	70.5	144.0	34.0	558	39
1067	0	66	77.7	144.0	37.5	533	53
1083	0	64	72.0	144.0	34.7	490	51
1039	0	69	68.0	144.0	32.8	520	57
1014	0	63	65.0	156.0	26.7	586	77
1011	0	80	61.8	157.0	25.1	608	66
1051	0	64	74.0	163.0	27.9	599	54
1062	0	85	60.0	139.5	30.8	662	41
1018	0	79	81.0	165.0	29.8	542	50
1061	0	61	63.0	142.0	31.2	642	55
1081	0	96	42.4	155.0	17.6	685	46
1026	0	67	77.0	151.0	33.8	522	51
1001	0	64	70.0	155.0	29.1	522	82
1043	0	69	69.8	148.5	31.7	576	46
1046	0	69	73.5	139.5	37.8	516	43
1085	0	63	88.0	146.0	41.3	514	47
1027	0	66	75.0	165.0	27.5	612	52
1047	0	79	93.9	146.3	43.9	491	49
1042	0	67	55.0	137.0	29.3	615	66
1037	0	84	67.7	137.0	36.1	495	49
1005	0	66	79.0	169.0	27.7	521	60
1009	0	60	95.0	159.0	37.6	461	55
1003	0	65	58.3	156.0	24.0	682	82
1013	0	68	86.0	163.0	32.4	545	60
1044	0	72	60.5	141.8	30.1	664	58
1038	0	70	55.6	139.0	28.8	630	64
1080	0	87	65.6	141.0	33.0	635	57
1025	0	61	72.0	173.0	24.1	640	58
1050	0	64	75.8	157.0	30.8	427	48
1045	0	66	61.2	148.5	27.8	613	53
3027	0	60	78.0	179.5	24.2	600	48
3029	0	64	79.1	156.0	32.5	502	49
1056	0	72	69.5	152.0	30.1	604	73
1053	0	60	105.8	165.0	38.9	529	61
1008	0	65	57.0	167.0	20.4	610	81
1071	0	71	48.0	161.0	18.5	606	66

Raw Data—Descriptive (Continued)

Subject ID	Sex	Age, (yrs)	Weight (kg)	Height (cm)	BMI	Resistance	Reactance
1041	0	64	57.0	146.0	26.7	557	59
1063	0	89	54.5	135.0	29.9	609	53
1066	0	60	72.7	148.5	33.0	539	59
1021	1	77	82.0	177.0	26.2	485	43
1015	1	69	82.0	191.0	22.5	457	37
1016	1	79	78.0	178.0	24.6	510	50
1002	1	79	85.0	175.0	27.8	449	73
1034	1	75	86.0	155.0	35.8	478	56
1004	1	76	93.2	177.0	29.7	474	70
1084	1	64	113.6	160.0	44.4	414	48
1020	1	67	91.0	173.0	30.4	510	53
1082	1	73	91.0	169.0	31.9	448	43
1022	1	87	64.0	163.0	24.1	465	45
1012	1	81	79.0	170.0	27.3	536	71
3001	1	63	114.9	199.0	29.0	436	42
3015	1	63	71.1	173.9	23.5	513	61
3005	1	67	84.2	179.7	26.1	462	46
3010	1	63	109.3	183.0	32.6	469	48
3008	1	66	109.8	178.1	34.6	444	47
3006	1	67	107.6	180.7	33.0	429	42
3017	1	60	83.9	177.3	26.7	456	53
3014	1	72	171.0	177.5	54.3	400	40
1052	1	89	60.0	160.0	23.4	475	34
1006	1	84	68.0	163.0	25.6	549	150
1036	1	61	95.0	184.0	28.1	466	52
1064	1	84	84.0	160.0	32.8	486	42
1055	1	76	78.7	178.0	24.8	462	45

Raw Data—FFM, %BF measured by ADP, BIA and DXA

Subject ID	BIA FFM	BIA %BF	BIA Absolute percent error%	ADP FFM	ADP %BF	ADP Absolute percent error	DXA FFM	DXA %BF
1019	46.9	35.8	4.4	46.6	35.2	2.6	47.8	34.3
1068	36.5	48.2	15.1	33.6	51.6	23.2	41.0	41.9
1067	40.9	47.4	12.0	40.8	47.7	12.8	45.3	42.3
1083	41.1	42.9	17.3	19.5	40.7	11.2	47.2	36.6
1039	39.7	41.7	22.9	41.7	39.3	15.9	46.8	33.9
1014	42.4	34.7	0.3	40.7	36.4	5.2	42.1	34.6
1011	39.8	35.7	13.2	33.3	46.0	11.9	38.0	41.1
1051	43.0	41.9	1.7	39.5	46.6	13.1	43.5	41.2
1062	30.3	49.5	37.8	34.0	40.1	11.7	38.7	35.9
1018	47.1	41.8	6.1	42.9	46.1	17.0	49.0	39.4
1061	33.9	46.2	10.9	38.6	46.6	11.8	39.9	41.7
1081	29.8	29.6	10.3	13.8	27.1	1.0	31.0	26.8
1026	42.9	44.2	1.7	41.8	45.1	3.7	43.4	43.5
1001	46.6	33.5	11.9	39.8	42.5	11.8	43.6	38.0
1043	37.8	45.8	19.3	37.3	46.4	20.8	44.0	38.4
1046	38.0	48.3	16.4	16.8	49.5	19.3	44.0	41.5
1085	43.8	50.2	20.1	21.3	45.8	9.6	52.8	41.8
1027	43.0	42.6	13.7	40.8	44.9	19.7	46.9	37.5
1047	46.5	50.5	14.2	48.1	48.8	10.4	53.3	44.2
1042	33.0	40.0	17.7	36.4	33.8	0.6	37.4	34.0
1037	37.6	44.6	14.8	35.1	48.0	23.7	43.2	38.8
1005	50.3	36.3	0.5	51.0	35.1	2.8	51.2	36.1
1009	53.4	43.8	7.6	47.5	50.0	5.5	49.6	47.4
1003	38.5	33.9	11.6	35.3	39.2	2.1	36.4	38.4
1013	48.8	43.2	11.7	48.8	42.9	10.9	53.5	38.7
1044	33.1	45.3	30.2	34.7	42.6	22.4	40.7	34.8
1038	33.0	40.8	12.3	36.7	33.9	6.6	36.3	36.3
1080	34.7	47.1	15.5	33.9	46.8	14.7	38.8	40.8
1025	44.3	38.5	18.8	47.3	32.7	0.9	48.8	32.4
1050	49.5	34.6	3.8	43.1	42.7	18.6	49.1	36.0
1045	35.6	41.9	36.0	17.9	35.6	15.6	44.0	30.8
3027	47.9	38.5	3.0	45.7	40.8	2.8	45.6	39.7
3029	45.7	42.3	2.6	45.5	41.8	1.5	45.4	41.2
1056	41.3	40.6	5.6	39.1	43.7	13.5	43.4	38.5
1053	54.9	48.1	4.8	54.8	48.1	4.8	57.5	45.9
1008	43.3	24.1	9.8	42.4	26.1	2.2	43.2	26.7
1071	37.7	21.4	1.5	41.0	14.3	32.2	38.6	21.1
1041	36.6	35.9	28.1	37.7	34.7	23.9	43.7	28.0

Raw Data—FFM, %BF measured by ADP, BIA and DXA (Continued)

Subject ID	BIA FFM	BIA %BF	BIA Absolute percent error	ADP FFM	ADP %BF	ADP Absolute percent error	DXA FFM	DXA %BF
1063	30.9	43.3	10.3	30.8	44.2	12.3	34.0	39.3
1066	41.6	42.8	13.0	39.9	45.2	19.2	46.0	37.9
1021	58.1	29.1	15.6	56.1	29.7	17.9	60.6	25.2
1015	65.2	20.5	30.3	72.5	9.9	36.9	69.3	15.7
1016	56.8	27.1	6.0	57.8	24.8	3.1	57.9	25.6
1002	64.6	24.0	1.4	63.5	25.1	5.9	67.5	23.7
1034	53.3	38.0	24.6	54.2	37.1	21.6	61.7	30.5
1004	65.0	30.3	1.2	64.5	30.9	3.3	68.3	29.9
1084	64.6	43.1	31.8	31.3	38.9	19.0	77.3	32.7
1020	58.4	35.8	22.1	58.2	35.0	19.5	64.7	29.3
1082	59.8	34.3	61.2	21.0	23.1	8.5	73.0	21.3
1022	50.4	21.3	6.6	52.7	17.5	12.5	52.8	20.0
1012	55.5	29.7	15.6	51.8	32.1	24.9	58.6	25.7
3001	79.2	31.1	1.2	79.8	30.7	0.0	76.6	30.7
3015	55.0	22.6	8.5	52.2	24.5	17.8	53.7	20.8
3005	61.8	26.6	0.6	60.2	28.5	8.0	61.2	26.4
3010	68.6	37.2	5.5	62.3	41.9	6.3	64.0	39.4
3008	68.6	37.5	4.4	65.5	40.4	12.5	69.3	35.9
3006	69.9	35.1	8.3	70.8	32.9	1.5	70.3	32.4
3017	62.1	25.9	0.2	57.5	30.2	16.2	59.5	26.0
3014	85.7	49.9	28.3	66.2	44.7	14.9	71.5	38.9
1052	46.3	22.8	6.6	50.5	15.6	36.1	47.0	24.4
1006	60.4	11.2	52.7	49.7	25.0	5.9	51.5	23.6
1036	66.5	30.0	0.8	62.5	34.1	12.5	66.4	30.3
1064	52.3	37.8	35.4	56.7	32.1	15.1	62.1	27.9
1055	59.7	24.2	27.2	64.4	18.0	5.3	65.0	19.0