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## Validity and reliability of a 4-compartment body composition model using dual energy x-ray absorptiometry-derived body volume

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## Abstract

**Background**—Body volume (BV), one component of a four-compartment (4C) body composition model, is commonly assessed using air displacement plethysmography (BodPod). However, dual-energy x-ray absorptiometry (DEXA) has been proposed as an alternative method for calculating BV.

**Aims**—This investigation evalauted the validity and reliability of DEXA-derived BV measurement and a DEXA-derived 4C model (DEXA-4C) for percent body fat (%BF), fat mass (FM), and lean mass (LM).

**Methods**—A total sample of 127 men and women (Mean  $\pm$  SD; Age: 35.8  $\pm$  9.4 years; Body Mass: 98.1  $\pm$  20.9 kg; Height: 176.3  $\pm$  9.2 cm) completed a traditional 4C body composition reference assessment. A DEXA-4C model was created by linearly regressing BodPod BV with DEXA FM, LM, and bone mineral content as independent factors. The DEXA-4C model was validated in a random sub-sample of 27 subjects. Reliability was evaluated in a sample of 40 subjects that underwent a second session of identical testing.

**Results**—When BV derived from DEXA was applied to a 4C model, there were no significant differences in %BF (p=0.404), FM (p=0.295), or LM (p=0.295) when compared to the traditional 4C model. The approach was also reliable; BV was not different between trials (p=0.170). For BV, %BF, FM, and LM relative consistency values ranged from 0.995-0.998. Standard error of

Conflict of Interest

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The authors have no conflicts to declare.

measurement for BV was 0.62L, ranging from 0.831-0.960kg. There were no significant differences between visits for %BF (p=0.075), FM (p=0.275), or LM (p=0.542).

**Conclusion**—The DEXA-4C model appears to be a valid and reliable method of estimating %BF, FM, and LM. The prediction of BV from DEXA simplifies the acquisition of 4C body composition by eliminating the need for an additional BV assessment.

#### Keywords

anthropometry; percent body fat; fat mass; lean mass; bioelectrical impedance

## INTRODUCTION

The science of body composition measurement is expanding as it plays an important role in disease detection and prevention. Excess fat mass has been associated with orthopedic injury, cardiovascular disease, and other indices of metabolic dysfunction [1-3].Conversely, inadequate lean mass and bone mineral content have been associated with increased musculoskeletal injury risk in aging and clinical populations, as well as compromised performance in athletes [4, 5]. Sophisticated anthropomorphic measures such as percent body fat (%BF), regional adiposity, and fat to lean mass ratio have been demonstrated as more suitable health predictors than the commonly used body mass index (BMI) [6, 7]. A variety of methods for assessing whole body composition have been developed to better evaluate each individual's health status, but technology is improving in order to better estimate body tissues.

Common body composition assessment techniques such as skinfold analysis and bioelectrical impedance are based on two-compartment (2C) models, which divide the body into fat mass (FM) and fat free mass (FFM). Such models assume uniform composition of FFM in making anthropomorphic predictions, despite the variation that exists in total body water (TBW), protein mass, and bone mineral content (BMC) [8, 9]. To compensate for such assumptions, multi-compartment models have been developed to individually assess the varying components of FFM [10]. A four-compartment (4C) model factoring in body mass (BM), body volume (BV), TBW, and BMC is considered by many as the gold standard in body composition [11].

The 4C model measurement, and associated body compartments, is accomplished using a variety of equipment, but also requires considerable time and cost. Dual energy x-ray absorptiometry (DEXA) is used to estimate total body BMC. The gold standard for TBW measurement is the use of deuterium oxide dilution; however, estimates have been shown to be valid when using multifrequency bioelectrical impedance spectroscopy (BIS).[12] Historically, underwater weighing (UWW) has been the standard method of determining BV based on hydrodensitometry. In recent decades, air displacement plethsmyography (ADP) has replaced UWW as a less invasive and more reliable method of assessing BV [13, 14]. Though considered more convenient than earlier methods, ADP requires specialized equipment (BodPod<sup>®</sup>), tight fitting clothing, and may be highly variable based on subject attire and body hair [15]. Additionally, both ADP and UWW must make assumptions

regarding trapped air in the digestive tract or lungs that may compromise validity in certain individuals [16].

Dual x-ray absorptiometry may serve as an alternative method of estimating BV [11, 16]. Unlike other displacement techniques, the x-ray attenuations utilized by DEXA exclude internal air voids when analyzing soft tissues, and therefore may provide more accurate volume estimations. One previous investigation from Wilson et al.[11] has suggested that DEXA may be used to determine BV, but the population utilized was small (n=11) and the authors suggested that further validation with a larger sample is necessary. The ability to use a DEXA-derived method for BV estimation may eliminate the need for ADP and/or UWW, reducing the time and cost required to obtain BV and use in a multi-compartment model. Greater testing efficiency would make use of a 4C body composition model more practical in both clinical and laboratory settings. Therefore, the aims of the current investigation were three-fold: 1) to develop a method of deriving BV from standard DEXA tissue measurements; 2) evaluate the validity of using DEXA-derived BV in a 4C body composition model; and 3) determine the reliability of DEXA-derived BV and 4C composition variables, including %BF, FM, and LM.

## MATERIALS and METHODS

#### **Participants**

A sample of 127 men and women (Mean  $\pm$  SD; Age: 35.8  $\pm$  9.4 years; Body Mass: 98.1  $\pm$  20.9 kg; Height: 176.3  $\pm$  9.2 cm, BMI: 31.4  $\pm$  5.5 kg·m<sup>-2</sup>) volunteered to participate in body composition assessments for two separate approved studies (IRB#12-1026, 14-1045). Participant BMIs ranged from normal to obese (BMI:19.9 –  $45.6 \text{ kg} \cdot \text{m}^{-2}$ ); with 104 Caucasians, 19 African Americans, and 3 Hispanics. A sample of 100 people was used to develop the coefficients reported in equation 2; a subsample of 27 was used to cross-validate the equation (Figure 1). All procedures were approved by the University's Biomedical Institutional Review Board. Prior to testing, all subjects reviewed and signed a written informed consent. In this observational study, subjects were excluded from the study if they were taking medication known to affect hydration status; if they were pregnant or lactating; or if they had undergone weight loss surgery. Subjects reported to the laboratory following a minimum of an eight-hour fast for a single body composition testing session. Additionally, subjects were asked to abstain from caffeine, alcohol, and vigorous exercise at least 24 hours prior to the testing session. To evaluate the reliability of testing methods, a sample of participants (n=40) from the original 127 underwent a second session of identical body composition testing at least 7 and no more than 10 days following their initial visit.

#### 4-compartment model

Percent body fat was estimated using a criterion 4C model described by Wang et al.[17]

$$FM (kg) = 2.748 (BV) - 0.699 (TBW) + 1.129 (Mo) - 2.051 (BM);$$
  
%BF = (FM/BM) × 100;  
FFM (kg) = BM - FM. Eq. 1

The model was calculated using traditional measurements of BV from ADP, TBW from bioelectrical impedance spectroscopy (BIS), and total body bone mineral (Mo) from DEXA. A modified 4C model was calculated to determine FM using a DEXA-derived value of BV based on mass measurements found on a standard whole body DEXA report.

#### **Bioelectrical Impedance Spectroscopy**

Total body water was determined using a multi-frequency BIS (SFB7, ImpediMed, Queensland, Australia) [12]. Estimates were taken after the subject lay supine for a minimum of 5 minutes. Two single tab electrodes were placed at the distal end of the subject's right wrist and hand and right ankle and foot, with 5 cm between each respective set of electrodes. Measurements were taken while the subject lay supine on the table with a space between their arms and torso and space between their legs. The average of two trials was used to represent TBW. The measurement of TBW from BIS has been shown to be valid against a deuterium dilution method [12]. Based on procedures described by Weir et al.[18], test-retest reliability (n=35) from our laboratory demonstrated intraclass correlation coefficients (model 2,1) were 0.99, standard error of measurement of 0.93 L (1.86% of the mean), with no systematic error between testing days (p=0.594).

#### Air displacement plethysmography

Body volume and BM were determined for the traditional equation from ADP using the BodPod<sup>®</sup>(Cosmed, USA Software V 4.2+), which was calibrated prior to each use according to the manufacturer's guidelines. The subject's weight (kg) was measured, while wearing a tight fitting bathing suit or compression shorts and sports bra, and swim cap, to the nearest 0.01 kg using the device's corresponding scale (Tanita Corp, Tokyo Japan). Two trials were performed for each subject to obtain two BV measurements within 150 mL. Thoracic lung volume was estimated, with previous reports demonstrating no differences between predicted and measured lung volumes [19, 20].

#### **Dual energy X-ray Absorptiometry**

Dual energy x-ray absorptiometry (Hologic, Discovery W, Bedford, MA) was used to estimate FM, LM, and total BMC using the device's default software (Apex V 3.3) for the original 4C model. This software version does not utilize the National Health and Nutrition Examination Survey (NHANES) correction factor [21]. Bone mineral content was further converted to Mo using the following equation: Mo = total body BMC  $\times$  1.0436. The device was calibrated in accordance with manufacturer guidelines before testing occurred to ensure valid results. Participants wore athletic-type clothing free of any metal and removed all jewelry prior to scanning. Participants were centered on the DEXA table in a supine position and instructed to lie still for the duration of the 7-minute scan. A trained researcher analyzed each scan to adjust software-determined regions of interest prior to producing the wholebody report. In order to determine body volume from DEXA, a prediction equation proposed by Wilson et al. was used [Eq. 2] [16]

DEXA Volume (L) = 
$$\frac{FM}{V_{Fat}} + \frac{LM}{V_{Lean}} + \frac{BMC}{V_{BMC}} + \nu_{Residual}$$
 Eq. 2.

where FM, LM, and BMC represent mass measurements acquired from DEXA. The coefficients correspond to the respective average densities for FM, LM, and BMC, while  $v_{\text{residual}}$  represents the volume of components unaccounted for by the DEXA.

#### **Statistical Analysis**

Using a sample of one hundred subjects, linear regression was performed using the enter method with BV from BodPod as the dependent variable, and FM, LM, and BMC from DEXA as independent factors, in order to determine the coefficients in Eq. 2. Using the new coefficients, DEXA BV was calculated in a randomly selected sub-sample of 27 subjects and applied to a 4C equation. The traditional 4C criterion model was compared to the DEXA-derived 4C model using dependent samples t-tests; constant error (CE = actual (4C) – predicted), total error (TE = [predicted-actual]<sup>2</sup>/n) and standard error of the estimate (SEE = [predicted-actual]• 1-r<sup>2</sup>) were calculated to assess the validity of %BF, FM, and LM. SPSS (IBM Corp, IBM SPSS Statistics for Windows, Version 21.0, Armonk, NY), was used to perform validity testing. In addition, Bland and Altman plots [22] were constructed to determine the 95% limits of agreement between the traditional 4C model and DEXA-4C model for BV, FM, %BF, and LM [23] (Figure 2).

Test re-test reliability for the DEXA-4C was evaluated in a randomly selected subset of the total sample (n=40) across two days using model "2,1" from Shrout and Fleiss [24] to determine intraclass correlation coefficient (ICC), standard error of measurement (SEM), and minimum difference (MD) values to be considered real [18]. Systematic variability between days was evaluated using a one-way repeated measures analysis of variance (ANOVA). Data were analyzed using a custom-written software program (Microsoft Excel, Microsoft Corporation, Redmond, WA, USA)

#### RESULTS

#### Validity

The inverse of the density coefficients of FM, LM, and BMC determined to predict BV were 0.84 (P<0.01), 1.03 (P<0.01), and 11.63 (P=0.853) respectively, with a residual volume of -3.12 L.

DEXA Volume (L) =  $\frac{FM}{0.84} + \frac{LM}{1.03} + \frac{BMC}{11.63} + (-3.12)$  Eq. 3.

Compared to the sub-sample, BV derived from DEXA (94.55  $\pm$  17.74 L) was not significantly different than BodPod BV (94.33  $\pm$  17.58 L; P=0.295, CI: [-1.05-0.01]). No significant differences were seen between any of the traditional predictions and the body composition variables calculated using the DEXA-4C criterion model; %BF (P =0.404, CI: [-2.61-0.25]); FM (P=0.295; CI: -2.91-0.02]); LM (P=0.295; CI: {-0.02-2.91]}). Constant error (CE), total error (TE), and the standard error of estimate (SEE) between the two models are presented in **Table 1**. Validity at the individual level using Bland-Altman plots are presented in Figure 2 for BV (1A), FM (1B), %BF (1C), and LM (1D).

## Reliability

Body volume derived from DEXA at visit 1 (Mean  $\pm$  SD; 92.96  $\pm$  17.54 L) was not significantly different from visit 2 (92.79  $\pm$  17.45 L; p=0.170) (Table 2). The relative consistency (ICC<sub>2,1</sub>) for BV was 0.998, whereas the SEM and MD values were 0.62 and 1.72 L, respectively. There were no significant differences between visits 1 and 2 for %BF ( 0.42  $\pm$  1.41%; p=0.075), FM ( 0.21  $\pm$ 1.17 kg; p=0.275), or LM ( -0.14  $\pm$  1.43 kg; p=0.542). The reliability statistics for body composition are presented in Table 2.

## DISCUSSION

Traditionally, the measurement of body composition using a 4C-model has required a BV measurement using either hydrostatic weighing or ADP and measurements of TBW from deuterium oxide. This multi-compartment measurement requires about 4 hours to complete for one participant, allowing for deuterium oxide to equilibrate, and is considered a gold standard [26]. The results of the current study demonstrate that a single DEXA scan may be used as an accurate method for determining BV and subsequent multi-compartment evaluation of body composition when combined with a more simplified and validated BIS assessment of TBW [12]. In the present study, a prediction model created from DEXA coefficients using LM, FM, and BMC [Eq. 2] provided a valid estimation of BV, compared to a traditional assessment from ADP. In the present study, the 4C model using DEXAestimated BV resulted in %BF, FM, and LM values not significantly different than those obtained from a traditional 4C-model utilizing ADP. The alternative method of predicting BV using this proposed equation from DEXA may allow for a valid multi-compartment evaluation of body composition without requiring additional equipment or testing time of ADP. This approach would only require a DEXA scan and measurement of TBW, significantly reducing subject burden and time constraints. Furthermore, the agreement of all body composition variables across two separate visits suggests good reliability for the DEXA-4C model. These findings contribute to a previous evaluation [16], including a larger, diverse population, thus enhancing generalizability.

## Accuracy

Underwater weighing was previously accepted as the gold standard for BV measurements; however, ADP has become the more acceptable approach for volume measurements [27]. Despite the widespread use of ADP for BV measurements, inaccuracies related to gastrointestinal gas and clothing may influence these results [28]. The use of DEXA to determine BV appears to be valid and eliminates inherent assumptions associated with displacement techniques [11]. In the present study, regressing LM, FM, and BMC resulted in an accurate BV estimation [Eq. 2]. The regression analysis demonstrated that FM and LM had slopes not significantly different from zero therefore contributing meaningful input toward volume estimations; whereas the slope coefficient for BMC was significantly different from zero. This suggests BMC has a non-significant impact on BV estimation equation [Eq. 2], but was maintained to provide a similar formula as previously reported [16]. Wilson et al.[16], who previously constructed unique BV coefficients, also demonstrated a slope for BMC that was significantly different from zero, which also yielded valid BV estimates. With low total error (1.06 L), high correlation (0.998), and low SEE

(0.07L), the generated coefficients from the present study accurately estimates BV in comparison to ADP. As a result of the coefficients being determined from a Hologic DEXA model, the formula may be limited to BV estimates from Hologic scans, which is a limitation to the current study. However, as an exploratory evaluation of this limitation, BV estimates using 30 subjects from our laboratory were scanned using a GE iDEXA were generated and evaluated, yielding similar validity to the current study. This is initial support that the equation can be generalized to various DEXA units, but future work should explore this utility.

For individual BV differences, the Bland and Altman plot demonstrated small differences with a slightly positive bias (0.29L) between ADP and DEXA-derived approach. There were also no significant mean differences for BV, suggesting that at a group or individual level body volume can be estimated from Eq. 2 using a single DEXA scan. Body volume has traditionally been assessed using ADP; body composition values derived from ADP alone have been shown to be imprecise for single 2C measurements, but with high reliability (14,16). The current findings suggest that DEXA-derived BV estimates may be used in place of ADP.

## Validity with the 4C ADP model

When utilizing DEXA-derived BV, the accuracy of the proposed DEXA-4C model was examined in comparison to the traditional ADP derived 4C model. On a whole group level, there were no significant mean differences for %BF, FM, or LM when using the DEXA-4C model (Table 2). In addition to similar mean values and high correlations (0.90-0.97), total error values ranged from 'good to very good' (TE = 3.07 - 3.40) [25], and standard error of the estimate values were considered ideal (Table 2). Individual analysis according to the Bland Altman plots produced larger limits of agreement than expected. For FM and %BF there was a slight positive bias suggesting that when using the DEXA-derived 4C model FM and %BF may be over predicted by 0.80 kg and 0.84%, respectively (Figure 2). Wilson et al. [16] reported a range of %BF bias of -1.62-4.92%, whereas in the current study our limits of agreement were wider at -5.33-7.01%. This bias may be a result of a more heterogeneous sample. In contrast, LM had a slight negative bias at -0.80 kg, resulting in slightly lower LM estimations when compared to the traditional 4C model. Despite being a 3C model, a wide range of precision has been reported for the DEXA [29, 30], resulting in growing popularity as a single method for body composition measurement. A multi-compartment approach to body composition, however, is still the preferred method and the most accurate [27]. Combining the use of DEXA as a method for BV, with TBW estimations, would significantly increase the accuracy of the measurement, while reducing subject burden for time, tight fitting clothing, and cost, compared to a traditional 4C model.

#### Consistency

In addition to establishing validity, there was no systematic error (p 0.075) between testing days for %BF, FM, or LM when using the 4C DEXA-derived model. Data from the current study demonstrates acceptable reproducibility for the modified 4C model for all body composition variables. Intraclass correlation coefficients were high (0.982) and SEM and MD values were low (Table 2). The SEM values in the present study are similar when

compared to previous reports evaluating the traditional 4C model, with between-day variability of ~ 1kg (FM) between test [31]. Overall, using DEXA-derived BV in a 4C model appears to be sensitive to changes in body composition, and appears to have similar consistency to the traditional 4C model from our laboratory (unpublished) where ICC are 0.988 and SEM averages 0.836 kg and 0.869% for FM, LM and %BF, respectively. The present results suggest a DEXA-derived 4C model, using DEXA and TBW measurements is highly reliable and may be a practical approach for tracking changes in body composition.

#### Conclusions

Valid and reliable body composition techniques are essential for detecting clinical conditions associated with both over- and under-fatness, overall health, injury prevention, and tracking changes from diet and exercise [2, 4, 30, 32-34]. Despite the known improvement in precision when using a multi-compartment model for body composition, many researchers, clinicians, and coaches still use single 2C techniques or DEXA (3C) as methods for determining body composition. The choice of measurement technique is often limited by cost, time, and equipment availability. The ability to utilize DEXA to determine BV allows for an easy 4-compartment calculation of body composition. A DEXA scan takes 6-13 minutes, dependent on model and the addition of a 5-minute TBW measurement (using BIS) allows for an accurate and rapid multi-compartment analysis of body composition to be made. The coefficients created in the current study expand upon the work by Wilson et al. [16] facilitating a quick and accurate estimation of BV. This BV estimation can be further used in a multi-compartment model to precisely and reliably estimate %BF, FM, and LM. Future studies should further extend the use of this formula in a variety of populations. Additionally, evaluating the current coefficients against values from other DEXA models with higher resolution may improve the accuracy.

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## Figure 2.

Differences between DEXA-4C model and Traditional model for **A**) body volume (mean difference [mdiff] = 0.29 L, limits of agreement [LOA] = -1.84 to 2.42 L); **B**) fat mass (mdiff = 0.80 kg, LOA = -5.06 to 6.65 kg); **C**) percent body fat (%BF; mdiff = 0.84%, LOA = -5.33 to 7.01 kg), and **D**) lean mass (mdiff = -0.80 kg, LOA = -6.65 to 5.06 kg).

#### Table 1

Constant error (CE), total error (TE), standard error of estimate (SEE) of BV, LM, %BF, and FM between traditional and the DEXA 4C model.

	BV (L)	%BF	FM (kg)	LM (kg)
Constant Error	-0.23	-0.56	-0.63	0.63
Total Error	1.06	3.40 <sup>#</sup>	3.07	3.07#
Standard Error of Estimate	0.07	1.48	1.04	0.70

According to Heyward et al.[25]

# classified as a good to very good;

 $^{\wedge}$  classified as ideal.

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## Table 2

Reliability statistics of body composition variables derived from the DEXA 4C model between visits 1 and 2.

	BV (L)	%BF	FM (kg)	LM (kg) <sup>221</sup>
Visit 1 (X ± SD)	$92.9 \pm 17.5$	$31.9\pm8.2$	$29.9 \pm 10.9$	$64.8 \pm 13.4$
$Visit \; 2 \; (X \pm SD)$	$92.8 \pm 17.5$	$31.5\pm8.5$	$29.7 \pm 10.9$	$64.9 \pm 13.1$
ICC <sub>2,1</sub>	0.998	0.982	0.995	0.996
SEM	0.624	0.960	0.831	0.999
MD	1.72	2.60	2.30	2.75