

# TOTAL BODY FAT ESTIMATIONS FROM SUBCUTANEOUS ADIPOSE TISSUE IN SARCOPENIC AND NON-SARCOPENIC MEN: A FOUR-COMPARTMENT MODEL APPROACH

Paul H. Falcone, Chih-Yin Tai, Laura R. Carson, Michael P. Kim, Eric R. Serrano, Jeffrey R. Stout, Abbie E. Smith, Kristina L. Kendall, David H. Fukuda, Joel T. Cramer, ML Rea, Sarah E. Moon, and Jordan R. Moon

Presented By : Paul Falcone - paul@musclepharm.com - MusclePharm Sports Science Research Institute

## ABSTRACT

Body composition changes due to aging can result in increased fat and decreased muscle mass. Decreases in muscle mass and increases in fat can lead to sarcopenia and an increased risk of injuries related to falls as well as a decrease in quality of living. However, the ability to estimate total body fat (%Fat) using simple subcutaneous adipose tissue (SAT) measurements is still unclear. **PURPOSE:** Generate and validate %Fat equations using SAT measurements. **METHODS:** Using a double-cross validation model, 124 men were separated in two groups maintaining similar sarcopenia numbers as well as age range (Group A: Age 72 +/- 6, 81.55 +/- 9.63 kg, 176.5 +/- 5.0 cm, 29.9 +/- 5.2 %Fat, 52.5% Sarcopenic; Group B: Age 71 +/- 5, 84.19 +/- 8.86 kg, 176.0 +/- 6.5 cm, 30.4 +/- 5.2 %Fat, 63.5% Sarcopenic). Subjects had their body fat measured using a four-compartment model (4C), including body volume via the BodPod, total body water via deuterium dilution, and bone mineral via dual-energy X-ray absorptiometry (DXA). SAT was measured using a B-Mode ultrasound (US). **RESULTS:** Using all seven US sites (triceps, biceps, abdominal, suprailiac, quadriceps, hamstrings, and calf) resulted in an r value of 0.62, a standard error of the estimate (SEE) of 4.09 %Fat and a total error (TE) of 4.13 %Fat. Using the three most correlated sites (biceps, chest, and quadriceps) improved prediction errors compared to all sites by producing an r value of 0.73, an SEE of 3.57 %Fat and a total error (TE) of 3.57 %Fat. Compared to the 4C model, DXA produced an r value of 0.88, an SEE of 2.47 %Fat and a TE of 2.93 %Fat. **CONCLUSIONS:** SAT measurements in elderly men can be used to predict total body fat. However, due to the large TE and SEE values in comparison to DXA, US does not appear to be a substitution for sophisticated lab methods. Still, US can be used with advanced lab methods to quantify both total body %Fat as well as SAT.

## INTRODUCTION

Changes in body composition can lead to sarcopenia (muscle loss), which increases the possibility of accidental falls in older adults, leading to potential hip fractures and other injuries (1,2). Along with a six-fold increase in government health care costs for the aged by 2040, hip fracture costs alone are projected to be six billion dollars in the year 2040 (3). More importantly, 20% of those with hip fractures will not be able to walk (4), and the average individual at 80 years old lacks the muscle capacity to rise unassisted from a chair (5). These muscular and injury-related limitations not only increase health-related costs but detrimentally affect quality of life, as well as the ability to perform ADLs (6). Therefore, due to the direct association between sarcopenia-related injuries and the subsequent effects on ADLs and quality of life, there exists a need for methods that identify the early onset of sarcopenia.

Sarcopenia can be caused by both a decrease in muscle mass and an increase in fat. However, the most accurate methods of measuring body composition are large, bulky devices that require a subject to visit a laboratory. A portable device that can accurately measure subcutaneous fat would be useful for diagnosing sarcopenia in the elderly in their own homes. Though equations have been validated for younger populations, a standard set of equations has not yet been determined for older individuals. Therefore, the purpose of this study is to validate new SAT equations using ultrasound to calculate %Fat in the elderly.

## EXPERIMENTAL DESIGN

All body composition assessments were performed on the same day following a twelve-hour fast (*ad libitum* water intake was allowed up to one hour prior to testing). Participants were instructed to avoid exercise for at least twenty-four hours prior to testing. Hydration status was determined using specific gravity via handheld refractometry (Model CLX-1, precision = 0.001 +/- 0.001, VEE GEE Scientific, Inc. Kirkland, Washington) prior to all body composition measurements. Specific gravity values indicated all subjects were properly hydrated during both pre- and post-testing sessions (>1.005, <1.030) (7,8).

One hundred and four healthy older (>65 yrs) men participated in the investigation. All subjects were Caucasians without medical implants or prostheses, considered non-obese (< 30 BMI), and without pacemakers.

## METHODS

### Sarcopenic Classification

SMI = total body muscle mass from single frequency bioimpedance: (TBMM / BM x 100) (6,9)  
TBMM = [(HT<sup>2</sup>/R) x 0.401 + Sex x 3.825 + Age x -0.071] + 5.102

Class I                      Class II  
SMI: 37 - 31% men        SMI: > 31% men  
SMI: 28 - 22% women    SMI: > 22% women

TBMM = Total body muscle mass, BM = Body mass in kg, HT = Height in cm, R = Bioimpedance Resistance, Sex: Men = 1, Women = 2.

### Bioelectrical Impedance Analysis (BIA)

Bioelectrical impedance analysis (BIA) was used to calculate total body muscle mass, as mentioned above (Imp<sup>TM</sup> DF50, ImpediMed Limited, Queensland, Australia). After resting in a supine position for 5 to 10 minutes, resistance and reactance were measured using a single 50kHz frequency while the subjects lay supine on a table with their arms ≥ 30 degrees away from their torso with their legs separated. Each pair of total body electrodes was connected by a non-conductive strip allowing for a distance of 5 cm between electrode centers. After hair removal and cleaning with alcohol, whole body electrodes were placed on the right side of the body. Electrodes were placed at the wrist (dorsal surface at the ulnar styloid process) and ankle (dorsal surface between the malleoli) with the connection strip and connected electrode 5 cm distal from the wrist and ankle. Skeletal muscle index (SMI) was calculated using the equation reported by Janssen et al. (6,9) and used to classify sarcopenia. Previous test retest scans of 11 men and women measured 24-48 hours apart resulted in an SEM = 8.91, ICC = 0.99 for Resistance.

### Ultrasound

Ultrasound was used at 3 (biceps, chest and quadriceps) or 7 (triceps, biceps, abdominal, suprailiac, quadriceps, hamstrings, and calf) sites to measure %fat (IntelaMetrix BX-2000, IntelaMetrix Inc., Livermore, CA). A thin layer of water-soluble gel was applied to the transducer, which was then applied to the skin site perpendicular to the skin. Without compressing the tissue, the transducer is slid back and forth along the skin so that the signal may be averaged by the device. The measurements are then included in the new equations listed below for both groups A and B.

### New Equations for Ultrasound

New Equations From Group A	New Equations From Group B
<b>SUM 3 DXA</b>	<b>SUM 3 DXA</b>
%Fat = 0.349267(Age)+0.325488(kg)+0.469910(SUM3)-0.076180(cm)-20.977768	%Fat = 0.196538(kg)+0.484680(SUM3)-0.210310(cm)-0.051242(Age)+42.522563
<b>SUM 3 4C</b>	<b>SUM 3 4C</b>
%Fat = 0.447745(Age)+0.287871(kg)+0.404046(SUM3)-0.141163(cm)-10.040965	%Fat = 0.153529(kg)+0.562713(SUM3)-0.150083(cm)+0.154896(Age)+20.013941
<b>SUM 7 DXA</b>	<b>SUM 7 DXA</b>
%Fat = 0.418404(Age)+0.385702(kg)+0.195575(SUM7)-0.115549(cm)-26.220273	%Fat = 0.318056(kg)+0.171981(SUM7)-0.381396(cm)-0.098197(Age)+65.103167
<b>SUM 7 4C</b>	<b>SUM 7 4C</b>
%Fat = 0.489505(Age)+0.348660(kg)+0.128872(SUM7)-0.173685(cm)-11.621047	%Fat = 0.308414(kg)+0.174599(SUM7)-0.343102(cm)+0.118744(Age)+44.467232



### Four-Compartment Model

Fat mass (FM, kg) was estimated using the 4C model described by (2) and converted to % fat using the following equations:

4C = 2.748 x BV - 0.699 x TBW + 1.129 x Mo - 2.051 x BM  
%fat = (FM / BM) x 100

BV = Body volume (L); TBW = Total body water (L); Mo = Total body bone mineral content (kg); BM = Body mass (kg); M = total-body mineral (kg).

### Body Volume (BV)

BV was measured using the BOD POD<sup>®</sup>. Before each test, the BOD POD<sup>®</sup> (BP) was calibrated according to the manufacturer's instructions with the chamber empty using a cylinder of known volume (49.558L). The subject, in a tight-fitting bathing suit and swimming cap only, then entered and sat in the fiberglass chamber. The BP was sealed, and the subject breathed normally for 20 seconds while body volume (Vb) was measured. After this, the subject was connected to a breathing tube internal to the system to measure thoracic gas volume (VTG). This value was used to correct Vb for VTG. All BP measurements were performed by a BOD POD<sup>®</sup>-certified investigator who had previously demonstrated a SEM of 0.3447 L on eleven men and women measured 24-48 hours apart.

Siri (6) 2C FM = 4.570 x BV - 4.50 x BM  
Brozek (1) 2C FM = 4.570 x BV - 4.142 x BM  
BV = Body volume (L); BM = Body mass (kg).



### Dual-Energy X-Ray Absorptiometry (DEXA)

Bone mineral content (BMC) was estimated using dual-energy X-ray absorptiometry (DXA) (software version 10.50.086, Lunar Prodigy Advance, Madison, WI). BMC was converted to total-body bone mineral (Mo) using the following equation:

Mo (kg) = total body BMC (kg) x 1.0436

Previous test retest scans of eleven men and women measured 24 - 48 hours apart for Mo produced a standard error of measurement (SEM) of 0.050 kg.



### Deuterium Oxide (D<sub>2</sub>O)

A D<sub>2</sub>O tracer was used to estimate TBW. Prior to D<sub>2</sub>O ingestion, urine samples were collected from all subjects. Subjects were instructed to void their bladder as much as possible. After voiding the bladder completely, subjects ingested ≈ 11 grams of D<sub>2</sub>O along with a 100ml rinse of deionized water. The exact amount of deuterium oxide ingested for each subject was recorded. After a 4-hour equilibration period restricting defecation, urination, and food and water ingestion, subjects were instructed to provide a post-urine sample. Within 30 minutes of collection, all urine samples were pipetted into cryogenic vials and stored at -80°C for later analysis. The urine-diluted D<sub>2</sub>O was analyzed in triplicate using an isotope-ratio mass spectrometer, and the isotope abundances in the urine were calculated following the method of (4). TBW was then calculated from the dilution of isotopic water and corrected for the exchange of deuterium with nonaqueous tissue (9).

## STUDY PARTICIPANTS

	Men (n=124)	
	Group A	Group B
Age (y)	72 +/- 6	71 +/- 5
Height (cm)	176.5 +/- 5.0	176.0 +/- 6.5
Weight (kg)	81.55 +/- 9.63	84.19 +/- 8.86
%Fat	29.9 +/- 5.2	30.4 +/- 5.2
%Sarcopenic	52.5	63.5

## RESULTS

DXA%FAT	(mean +/- SD) (%)	Slope	Intercept	r	SEE (%)	TE (%)	Agreement			
							CE (%) / Bias +/- 2SD	Upper Limits	Lower Limits	Trend
<b>Men A (n = 61)</b>										
<b>4C</b>	<b>29.92 +/- 5.16</b>									
<b>SUM 3 4C</b>	<b>30.07 +/- 3.46</b>	<b>0.91</b>	<b>2.41</b>	<b>0.61</b>	<b>4.11</b>	<b>4.05</b>	<b>-0.16 +/- 8.00</b>	<b>7.84</b>	<b>-8.16</b>	<b>0.48</b>
<b>SUM 7 4C</b>	<b>29.32 +/- 3.34</b>	<b>0.83</b>	<b>5.66</b>	<b>0.54</b>	<b>4.39</b>	<b>4.40</b>	<b>0.60 +/- 8.61</b>	<b>9.21</b>	<b>-8.01</b>	<b>0.55</b>
<b>DXA % Fat</b>	<b>27.95 +/- 5.67</b>	<b>0.81</b>	<b>7.20</b>	<b>0.89</b>	<b>2.34</b>	<b>3.20</b>	<b>1.96 +/- 5.00</b>	<b>6.96</b>	<b>-3.03</b>	<b>-0.10</b>
<b>SUM 3 DXA</b>	<b>28.90 +/- 3.47</b>	<b>1.01</b>	<b>-1.35</b>	<b>0.62</b>	<b>4.48</b>	<b>4.50</b>	<b>-0.95 +/- 8.70</b>	<b>7.76</b>	<b>-9.65</b>	<b>0.58</b>
<b>SUM 7 DXA</b>	<b>28.25 +/- 3.78</b>	<b>0.89</b>	<b>2.80</b>	<b>0.59</b>	<b>4.60</b>	<b>4.55</b>	<b>-0.30 +/- 8.97</b>	<b>8.67</b>	<b>-9.27</b>	<b>0.50</b>
<b>Men B (n = 63)</b>										
<b>4C</b>	<b>30.39 +/- 5.24</b>									
<b>SUM 3 4C</b>	<b>30.48 +/- 4.79</b>	<b>0.89</b>	<b>3.14</b>	<b>0.82</b>	<b>3.03</b>	<b>3.03</b>	<b>-0.09 +/- 5.98</b>	<b>5.89</b>	<b>-6.06</b>	<b>0.10</b>
<b>SUM 7 4C</b>	<b>30.80 +/- 4.41</b>	<b>0.82</b>	<b>5.11</b>	<b>0.69</b>	<b>3.81</b>	<b>3.85</b>	<b>-0.41 +/- 7.57</b>	<b>7.16</b>	<b>-7.98</b>	<b>0.20</b>
<b>DXA % Fat</b>	<b>29.47 +/- 4.88</b>	<b>0.94</b>	<b>2.56</b>	<b>0.88</b>	<b>2.51</b>	<b>2.65</b>	<b>0.92 +/- 4.91</b>	<b>5.83</b>	<b>-3.99</b>	<b>0.08</b>
<b>SUM 3 DXA</b>	<b>28.61 +/- 5.19</b>	<b>0.74</b>	<b>8.36</b>	<b>0.79</b>	<b>3.04</b>	<b>3.40</b>	<b>0.86 +/- 6.49</b>	<b>7.35</b>	<b>-5.63</b>	<b>-0.07</b>
<b>SUM 7 DXA</b>	<b>28.98 +/- 5.16</b>	<b>0.63</b>	<b>11.27</b>	<b>0.66</b>	<b>3.68</b>	<b>4.12</b>	<b>0.49 +/- 8.07</b>	<b>8.56</b>	<b>-7.59</b>	<b>-0.07</b>
<b>Men A+B (n = 124)</b>										
<b>4C</b>	<b>30.16 +/- 5.18</b>									
<b>SUM 3 4C</b>	<b>30.28 +/- 4.18</b>	<b>0.90</b>	<b>2.86</b>	<b>0.73</b>	<b>3.57</b>	<b>3.57</b>	<b>-0.12 +/- 7.02</b>	<b>6.89</b>	<b>-7.14</b>	<b>0.25</b>
<b>SUM 7 4C</b>	<b>30.07 +/- 3.98</b>	<b>0.81</b>	<b>5.94</b>	<b>0.62</b>	<b>4.09</b>	<b>4.13</b>	<b>0.09 +/- 8.13</b>	<b>8.21</b>	<b>-8.04</b>	<b>0.32</b>
<b>DXA %fat</b>	<b>28.72 +/- 5.31</b>	<b>0.86</b>	<b>5.50</b>	<b>0.88</b>	<b>2.47</b>	<b>2.93</b>	<b>1.43 +/- 5.04</b>	<b>6.47</b>	<b>-3.60</b>	<b>-0.03</b>
<b>SUM 3 DXA</b>	<b>28.75 +/- 4.41</b>	<b>0.81</b>	<b>5.30</b>	<b>0.68</b>	<b>3.93</b>	<b>3.98</b>	<b>-0.03 +/- 7.83</b>	<b>7.80</b>	<b>-7.86</b>	<b>0.22</b>
<b>SUM 7 DXA</b>	<b>28.62 +/- 4.53</b>	<b>0.73</b>	<b>7.93</b>	<b>0.62</b>	<b>4.19</b>	<b>4.33</b>	<b>0.10 +/- 8.53</b>	<b>8.63</b>	<b>-8.43</b>	<b>0.20</b>

## CONCLUSIONS

Due to the large TE and SEE values, ultrasound cannot be considered as an alternative to more comprehensive lab methods like DXA in sarcopenic and non-sarcopenic elderly men. However, ultrasound can be used in conjunction with other lab methods to quantify subcutaneous adipose tissue as well as %Fat. Future research should evaluate SAT and its usefulness in overall health and performance. This study was funded by a grant provided by Abbott Nutrition:

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