Determination of skeletal muscle and fat-free mass by nuclear and dual-energy X-ray absorptiometry methods in men and women aged 51–84 y

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ABSTRACT
Background: Skeletal muscle mass (SMM) and fat-free mass (FFM) are important variables in nutritional studies. Accurate techniques for measuring these variables have not been thoroughly validated in elderly subjects.

Objectives: The objectives of this study were to 1) compare SMM values derived from dual-energy X-ray absorptiometry (DXA) with those calculated by a nuclear method from total body potassium (TBK) and total body nitrogen (TBN) measurement (both: KN) in older subjects, and 2) assess the accuracy of FFM measurement by DXA in these subjects.

Design: TBK, TBN, DXA (model XR36; Norland, Fort Atkinson, WI), bioimpedance, and anthropometric measurements were performed on healthy women (n = 50) and men (n = 25) aged 51–84 y.

Results: Mean SMM by KN was not significantly different from SMM by DXA in either sex. SMM by KN predicted SMM by DXA with an SEE of 2.1 kg (r = 0.95, P < 0.0001 for women and men together). In the men, FFM by DXA agreed well with FFM estimated by TBK, skinfold thicknesses, bioimpedance analysis, and a multicompartment model. In women, FFM by DXA was 4–5 kg less than that by the other methods (P < 0.01). Truncal fat was related to intermethod FFM differences (r = 0.58, P < 0.0001).

Conclusions: These data indicate that 1) the nuclear or the DXA method can be applied to estimate SMM in healthy older subjects, and 2) the Norland DXA instrument significantly underestimates FFM in older women, in part, because of the influence of truncal adiposity. Am J Clin Nutr 1999;70:228–33.

KEY WORDS Skeletal muscle mass, dual-energy X-ray absorptiometry, sarcopenia, total body potassium, total body nitrogen, bioimpedance analysis, aging, anthropometry, humans

INTRODUCTION
Skeletal muscle mass (SMM) is an important variable to consider in nutritional studies. Skeletal muscle is metabolically active, represents a large proportion of the fat-free mass (FFM) of the body, and should be maintained in the elderly to prevent frailty and loss of independence (1, 2). Although several studies have implied that a substantial loss of SMM (sarcopenia) is an inevitable feature of human aging (3–6), some evidence suggests that sarcopenia can be considerably minimized, and even reversed, by appropriate physical activity (1, 2, 7–9).

To quantify age-associated decreases in SMM and the effects of interventions, accurate estimation of SMM is required. This has proven to be difficult because there is no direct in vivo means of measuring SMM. There are, however, several methods of indirect estimation, including anthropometric fractionation (10); creatinine excretion (11); whole-body counting and neutron activation to quantify total body potassium (TBK) and total body nitrogen (TBN), respectively (12, 13); computed tomography (14); and magnetic resonance imaging (15). These methods are all time-consuming and technically difficult to perform, and those methods often ranked the highest for accuracy (computed tomography and magnetic resonance imaging) involve considerable radiation exposure or expensive instrumentation.

Dual-energy X-ray absorptiometry (DXA) is a relatively new method of body-composition analysis that involves minimal radiation (16). A whole-body DXA scan divides the body into bone, fat, and lean compartments. With appropriate definition of arm and leg regions, DXA provides an estimation of the fat-free soft tissue (FFST) in the limbs. If it is assumed that this limb FFST value closely represents limb SMM, as discussed by Heymsfield et al (17), then total SMM can be calculated for normal individuals on the basis of the proportion of limb SMM to total SMM (0.75) reported in cadaver studies (18). This method of SMM estimation was shown to agree well with computed tomography–determined SMM in 25 young to middle-aged men (13). In addition, DXA-determined limb SMM was found to correlate strongly with TBK in a sample of 148 women and 136 men aged 20–90 y (6). Because DXA can also quantify whole-body bone mineral density and bone mineral content (BMC) together with total FFM in a 5–6-min...
scan involving negligible radiation exposure (<0.05 mSv), it is emerging as a popular body-composition assessment tool.

DXA has not been thoroughly validated for SMM estimation in older subjects. The primary aim of this study was, therefore, to compare DXA-derived SMM values with those obtained via TBK and TBN measurement in a sample of 51–84-y-old women and men. Because some uncertainties exist regarding the accuracy of DXA determination of soft tissue components in older age groups (19–21), and few reports on the use of DXA in older age groups (19–21), and few reports on the use of DXA to compare DXA-derived SMM values with those obtained via TBK and TBN measurement in a sample of 51–84-y-old women and men. Because some uncertainties exist regarding the accuracy of DXA determination of soft tissue components in older age groups (19–21), and few reports on the use of DXA to compare DXA-derived SMM values with those obtained via TBK and TBN measurement in a sample of 51–84-y-old women and men. Because some uncertainties exist regarding the accuracy of DXA determination of soft tissue components in older age groups (19–21), and few reports on the use of DXA to compare DXA-derived SMM values with those obtained via TBK and TBN measurement in a sample of 51–84-y-old women and men. Because some uncertainties exist regarding the accuracy of DXA determination of soft tissue components in older age groups (19–21), and few reports on the use of DXA methods, they were all, by self-report, weight stable and apparently healthy. Each subject gave informed consent for the study, which was approved by the Royal North Shore Hospital Medical Research Ethics Committee and Radiation Protection Committee.

**SUBJECTS AND METHODS**

**Subjects**

The study sample comprised white women aged 54–84 y (n = 50) and white men aged 51–76 y (n = 25). The subjects had a wide range of body sizes and adiposity (Table 1). On recruitment, they were all, by self-report, weight stable and apparently healthy. Each subject gave informed consent for the study, which was approved by the Royal North Shore Hospital Medical Research Ethics Committee and Radiation Protection Committee.

**Body-composition measurements**

**Total body potassium and total body nitrogen**

TBK was measured by supine sodium iodide counting, as described previously (22). The precision and accuracy of this method, expressed as CVs, are 1.5% and 4.5%, respectively. TBK was used to estimate FFM (FFM_{TBK}), assuming that the potassium content of FFM is 2.26 g/kg in women and 2.52 g/kg in men (23). TBN was measured by in vivo neutron-capture analysis, as described by Allen et al (24), with a precision and accuracy of 3% and 4.5%, respectively. The radiation exposure with a TBN scan involving negligible radiation exposure (<0.05 mSv), it is emerging as a popular body-composition assessment tool.

**Anthropometry**

Height was measured to the nearest 0.5 cm with a wall-mounted stadiometer and body weight was measured to the nearest 0.1 kg with digital scales. Skinfold thicknesses were measured in duplicate by one researcher with constant-pressure calipers (Holtain Ltd, Crymych, United Kingdom) at the triceps, biceps, subscapular, and suprailiac sites. The skinfold-thickness measurements were used to estimate percentage body fat with the appropriate age- and sex-specific equations of Durnin and Womersley (27). The precision of percentage fat estimation was 1.1%. Skinfold thickness–derived FFM (FFM_{SKF}) was calculated from percentage body fat and body weight measurements.

**Bioimpedance analysis**

A bioimpedance analysis (BIA) measurement was taken after each subject had rested supine for 5 min, with electrodes in a tetrapolar configuration, by using a swept-frequency instrument (SEAC model SFB2.3 with associated software; UniQuest, Queensland, Australia). The BIA output measures were used to derive FFM (FFM_{BIA}) by applying the equation of Lukaski et al (28), and total body water (TBW) by using the equation of Kushner and Schoeller (29). The latter equation was derived in a group of men and women aged ≈20–70 y; the equation predicts deuterium oxide space from a combination of subject resistance, height, and weight. The precisions of FFM_{BIA} and TBW measurements were 1.6% and 1.4%, respectively.

**Data reduction and analysis**

**Skeletal muscle mass estimation**

SMM was estimated from TBK and TBN data (ie, SMM_{KN}) by using the equation of Wang et al (13) as follows:

\[
SMM_{KN} = (0.188 \times TBK) + (0.00183 \times TBN) \tag{1}
\]

where TBK and TBN are in grams and SMM_{KN} is in kilograms.

Because this equation was developed empirically by relating TBK and TBN to computed tomography–determined SMM in a multiple regression model, it can be regarded as a surrogate measure of computed tomography–determined SMM and should therefore accurately represent SMM.

DXA-derived SMM (SMM_{DXA} in kg) was calculated from the sum of arm and leg FFST values (in kg), assuming that this sum represents limb SMM and that limb SMM represents 75% of total body SMM, as discussed above:

\[
SMM_{DXA} = 1.333 \times (\text{arm FFST} + \text{leg FFST}) \tag{2}
\]
Table 2
SMM and FFM values by several methods

<table>
<thead>
<tr>
<th></th>
<th>Women</th>
<th>Men</th>
</tr>
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<tbody>
<tr>
<td>SMM&lt;sub&gt;KN&lt;/sub&gt;</td>
<td>19.9 ± 3.3</td>
<td>31.7 ± 4.0</td>
</tr>
<tr>
<td>SMM&lt;sub&gt;DXA&lt;/sub&gt;</td>
<td>19.6 ± 3.6</td>
<td>31.4 ± 4.4</td>
</tr>
<tr>
<td>FFM&lt;sub&gt;DXA&lt;/sub&gt;</td>
<td>37.4 ± 5.9</td>
<td>57.9 ± 6.7</td>
</tr>
<tr>
<td>FFM&lt;sub&gt;TBK&lt;/sub&gt;</td>
<td>41.1 ± 7.1</td>
<td>58.8 ± 7.9</td>
</tr>
<tr>
<td>FFM&lt;sub&gt;SKF&lt;/sub&gt;</td>
<td>41.3 ± 5.1</td>
<td>57.6 ± 6.4</td>
</tr>
<tr>
<td>FFM&lt;sub&gt;TK&lt;/sub&gt;</td>
<td>41.6 ± 5.3</td>
<td>57.3 ± 7.0</td>
</tr>
<tr>
<td>FFM&lt;sub&gt;4C&lt;/sub&gt;</td>
<td>42.6 ± 4.6</td>
<td>59.4 ± 6.6</td>
</tr>
</tbody>
</table>

<sup>1</sup> ± SD. SMM, skeletal muscle mass; FFM, fat-free mass; KN, total body potassium and nitrogen; DXA, dual-energy X-ray absorptiometry; TBK, total body potassium; SKF, skinfold thicknesses; BIA, bioimpedance analysis; 4C, 4-compartment model.

<sup>2</sup> Significantly different from men, P < 0.001 (Student’s t test for unpaired data).

<sup>3</sup> Significantly different from FFM<sub>DXA</sub>, P < 0.01 (Student’s t test for paired data).

Fat-free mass based on a 4-compartment body-composition model

A widely used 4-compartment body-composition model assumes that the body consists of fat, protein, water, and mineral compartments (30, 31). FFM (FFM<sub>4C</sub>, in kg) was calculated from this model as follows:

\[ \text{FFM}_{4C} = \text{protein} + \text{TBW} + \text{mineral} \] (3)

where protein is total body protein [TBN (in kg) × 6.25], TBW is determined by BIA (in kg), and mineral is total body mineral [total BMC (in kg)/0.84].

As discussed by Baumgartner (30), the assumed nitrogen content of protein (1 g N = 6.25 g protein) and the ratio of osseous to nonosseous mineral (0.84) involved in these calculations are applicable to subjects in this age range.

![Figure 1](image-url)  
**Figure 1.** Skeletal muscle mass determined by dual-energy X-ray absorptiometry (SMM<sub>DXA</sub>) versus that determined by the nuclear method (SMM<sub>KN</sub>) for women (○) and men (●) combined. The fine diagonal line is the line of identity; the bold line is the regression line.

Bone mineral and water content of fat-free mass

The mineral content of FFM was calculated by expressing BMC as a percentage of FFM<sub>SKF</sub>. Similarly, hydration of FFM was estimated by expressing TBW as a percentage of FFM<sub>SKF</sub>. These measures were included to assess their potential contribution to intermethod differences in body-composition measurement because variance in these FFM components is known to affect body density and can thereby reduce the accuracy of 2-compartment models (26, 30, 31).

Statistical analysis

Correlation and regression analysis, Student’s t tests, and Bland-Altman analyses (32) were used to compare the 2 methods of SMM estimation and to compare FFM<sub>DXA</sub> with the other methods (TBK, SKF, BIA, and 4C) of FFM determination. All statistical analyses were carried out with SPSS for WINDOWS (release 6.1.4; SPSS Inc, Chicago). The level of significance was set at P < 0.05 for all analyses.

RESULTS

Skeletal muscle mass estimates

Data for women and men analyzed separately

SMM<sub>DXA</sub> and SMM<sub>KN</sub> were significantly correlated in both the women (r = 0.83) and men (r = 0.87) (P < 0.0001 for both). Regression analysis showed that the 2 SMM methods were related as follows:

Women: SMM<sub>DXA</sub> = 0.90 SMM<sub>KN</sub> + 1.7  
(4)

Men: SMM<sub>DXA</sub> = 0.97 SMM<sub>KN</sub> + 0.5  
(5)

where SMM<sub>KN</sub> predicted SMM<sub>DXA</sub> with an SEE of 2.0 kg in the women and 2.2 kg in the men. The 95% CIs for the regression slopes in equations 4 and 5 include the line of identity.

Paired t tests showed no significant differences between the 2 estimates of SMM in data from women and men (Table 2). Bland-Altman analyses (32) did not show any systematic bias, for either sex, in the differences between the 2 methods as SMM values increased.

Pooled data

Because neither the slopes nor the intercepts of equations 4 and 5 were significantly different between the sexes, the data were pooled. In this combined data set, the 2 estimates of SMM were highly correlated and in good agreement, such that SMM<sub>KN</sub> predicted SMM<sub>DXA</sub> with an SEE of 2.1 kg (Figure 1). The 95% CIs for the regression slope given in Figure 1 include the line of identity. A paired t test showed that although mean SMM<sub>KN</sub> was 0.32 kg higher than mean SMM<sub>DXA</sub>, this difference was not significant. Further analysis with the Bland-Altman method (32) revealed that there was no systematic bias in the differences between the 2 methods as SMM values increased (Figure 2).

Age effects

Both SMM estimates correlated significantly and negatively with age for women but not for men. For women, SMM<sub>KN</sub> had a more significant relation with age (r = −0.42, P = 0.002) than did SMM<sub>DXA</sub> (r = −0.3, P = 0.035).
Mineral and water content of fat-free mass

Mean (±SD) values for BMC as a percentage of FFM were 5.4 ± 0.6% and 6.2 ± 0.8% for men and women, respectively. Mean values for TBW as a percentage of FFM were 73.0 ± 5.2% and 73.8 ± 4.3% in men and women, respectively.

Fat-free mass estimated by dual-energy X-ray absorptiometry compared with other methods

In contrast with the close agreement between SMM estimates, there were relatively large and significant differences between FFMDXA and several other FFM estimates, particularly in the data set from women (Table 2). To determine whether these intermethod differences were related to variables such as age, adiposity, fat distribution, BMC as a percentage of FFM, or TBW as a percentage of FFM, a series of Bland-Altman analyses (32) were performed on pooled data from men and women by plotting the difference between FFM4C and FFM DXA against these variables. The difference between the FFM methods was not related to age or percentage fat, but was related to BMC as a percentage of FFM (r = 0.43, P = 0.0001) and TBW as a percentage of FFM (r = 0.26, P = 0.02). There was a highly significant relation (r = 0.58, P < 0.0001) to truncal adiposity, as reflected in the ratio of truncal fat to leg fat (Figure 3).

DISCUSSION

The average values for weight, percentage fat, and bone mineral and water contents of FFM in these subjects were in close agreement with values for older white persons reported elsewhere. Snead et al (21) found that healthy women aged 60–73 y had a mean weight of 65 kg and a mean percentage fat (by hydrodensitometry) of 39%; values for men aged 60–82 y were 77 kg and 26%, respectively. Dual-photon absorptiometry data from Mazess et al (33) give a mean BMC:FFM value of 5.9% for women aged 50–61 y. Baumgartner (30) reported FFM hydration values of 74.3% and 71.0% in elderly women and men, respectively. Because the TBW values in the current study were obtained with a BIA technique, they should be interpreted with caution. Nevertheless, given the favorable comparisons summarized above, we expect that the subjects in the current study were representative of healthy older whites.

The strength of agreement between the SMMDXA and SMMKN estimates was striking, given that these methods involve totally independent assumptions. Although neither of the methods has been extensively validated against a gold standard SMM measure in older subjects, the close agreement between the regression line and the line of identity in Figure 1 implies that both methods were in fact measuring the same variable: SMM. Furthermore, because the equation used in this study to derive SMMKN was empirically derived from computed tomography scans in subjects with a considerably wide range of SMM (13), similar results would be expected if the DXA method were to be compared directly with computed tomography–derived SMM in healthy older subjects.

This agreement between the DXA and the KN methods implies that reasonably accurate determinations of SMM in healthy elderly people are possible by either method. The speed of data acquisition (5–6 min for the Norland model XR36 whole-body scan) and the lower radiation exposure with DXA compared with a TBN measurement (TBK involves no radiation exposure) make DXA an attractive option. In our laboratory, a combined TBN and TBK assessment takes ≈60 min to complete. However, the fact that we found SMMKN values to be more highly correlated with age than were SMMDXA values (in women), considered together with the lower SDs found in the SMMKN data (Table 2), could indicate advantages for the nuclear method.

Use of the nuclear techniques can yield valuable clinical information in addition to SMM estimation. Total body protein, assessed via TBN, is an important indicator of nutritional status and has been shown to reflect the severity of several illnesses (23, 24, 34). Comparison of patient values with age- and sex-matched norms from a healthy reference population can therefore be an important prognostic guide and assist in clinical decision-making.
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weight data from Snead et al. (21) shows that FFM\textsubscript{DXA} measure-
particularly in women, implying a considerable underestimation
were consistently higher than hydrodensitometry-derived values,
densitometry in 219 adult subjects. DXA-derived values
compared percentage fat values obtained by DXA (Norland) and
crepancies between FFM\textsubscript{DXA} and other FFM measures, particu-
measurement, we found significant dis-
tional assessment, particularly when serial changes associated
TBN contributes much valuable information to a patient’s nutri-
acute disturbances in nutritional status, severity of disease, and
show short-term changes, TBK is invaluable in the assessment of
content of FFM has been shown to decline with age (36), the
First, if the BMC and water content of FFM vary markedly
could potentially influence the intermethod FFM agreement.
Our finding that truncal adiposity was positively related to
intermethod FFM differences suggests that the fat distribution
model in the DXA system software is a critical factor in deter-
mucular techniques is recommended for thorough body-composi-
these conclusions apply only to healthy subjects. The use of
DXA should be used cautiously in the measurement of
factors, DXA should be used cautiously in the measurement of
In summary, the results of this study indicated that both DXA
and KN measurement can be used to estimate SMM in healthy
elderly subjects with reasonable accuracy and precision. DXA has
the practical advantage of convenience over the nuclear method.
However, when comprehensive body-composition assessment is
justified, particularly in the study of disease states in which con-
stancy assumptions are challenged, measurements of TBK and
TBN provide more detailed information for nutritional analysis.
Although DXA provides FFM values that agree quite well with
FFM from other methods in older men, it underestimates
FFM in older women, possibly because of inaccuracies in defining
truncal fat. Further studies are necessary to confirm whether other
DXA instrument models and software can provide accurate
assessment of SMM in this age group and in patient groups. Thus,
these conclusions apply only to healthy subjects. The use of
nuclear techniques is recommended for thorough body-composi-
tion investigations of disease states.

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REFERENCES

4. Flynn MA, Nolph GB, Baker AS, Martin WM, Krause G. Total body
8. Giavarone MA, Marks EC, Ryan ND, Meredith CN, Lipsitz L, Evans WJ. High-intensity strength training in nonagenarians. Effects on
9. Frontera WR, Meredith CN, O’Reilly KP, Knutigen HG, Evans WJ. Strength conditioning in older men: skeletal muscle hypertrophy and
11. Webster J, Garrow JS. Creatinine excretion over 24 hours as a measure of
12. Burkshinshaw L, Hill GL, Morgan DB. Assessment of the distribution of
tal body composition technique based on computerized tomography.
15. Engstrom CM, Loeb GE, Reid JR, Forrest WJ, Avruch L. Mor-
phometry of the human thigh muscles: a comparison between
anatomical sections and computer tomographic and magnetic reso-
16. Mazess RB, Barden HS, Bisek JP, Hanson J. Dual-energy x-ray
absorptiometry for total- body and regional bone-mineral and soft-
mass: measurement by dual-photon absorptiometry. Am J Clin
Task Group on Reference Man. Oxford, United Kingdom: Perga-
momon, 1975. (ICRP publication no. 23.)
DEXA in older adults: accuracy and influence of scan mode. Med
21. Sneed DB, Birge SJ, Kohrt WM. Age-related differences in body
composition by hydrodensitometry and dual-energy x-ray absorp-
22. Hansen RD, Allen BJ. Calibration of a total body potassium monitor
composition of cancer patients by measurement of total body nitro-
protein as a prognostic indicator in wasting disease. Asia Pac J Clin
25. Goodis MM. Evaluation of a new set of calibration standards for
the measurement of fat content via DPA and DXA. Med Phys
1992;19:35–44.
27. Durnin JV, Womersley J. Body fat assessed by total body density
28. Lukaski HC, Bolonchuk WW, Hall CB, Siders WA. Validation of
tetrapolar bioelectrical impedance method to assess human body
29. Kushner RF, Schoeller DA. Estimation of total body water by bio-
30. Baumgartner RN. Body composition in elderly persons: a critical
31. Lohman TG. Advances in body composition assessment. Cham-
32. Bland JM, Altman DG. Statistical methods for assessing agreement
33. Mazess RB, Peppler WW, Gibbons M. Total body composition by
34. Hill GL. Body composition research: implications for the practice of
35. Pollock CA, Ibels LS, Allen BJ, et al. Total body nitrogen as a prog-
36. Kehayias JJ, Fiatarone MA, Zhuang H, Roubenoff R. Total body potas-
37. Roche AF, Guo S, Wellens R, Chumlea WC, Wu X, Siervogel RM.
Fat-free mass from dual-energy x-ray absorptiometry and from
38. Nord RH, Payne RK. A new equation set for converting body density
39. Wellens R, Chumlea WC, Guo S, Roche AF, Reo NV, Siervogel RM.
Body composition in white adults by dual-energy x-ray absorptiometry,