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Four-component model for the assessment of body composition in humans: comparison with alternative methods, and evaluation of the density and hydration of fat-free mass

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1. Body composition was assessed in 28 healthy subjects (body mass index 20–28 kg/m^2) by dual-energy X-ray absorptiometry, deuterium dilution, densitometry, ^{40}K counting and four prediction methods (skinfold thickness, bioelectrical impedance, near-i.r. interactance and body mass index). Three- and four-component models of body composition were constructed from combinations of the reference methods. The results of all methods were compared. Precision was evaluated by analysis of propagation of errors. The density and hydration fraction of the fat-free mass were determined.

2. From the precision of the basic measurements, the propagation of errors for the estimation of fat (± SD) by the four-component model was found to be ± 0.54 kg, by the three-component model, ± 0.49 kg, by deuterium dilution, ± 0.62 kg, and by densitometry, ± 0.78 kg. Precision for the measurement of the density and hydration fraction of fat-free mass was ± 0.0020 kg/l and ± 0.0066, respectively.

3. The agreement between reference methods was generally better than between reference and alternative methods. Dual-energy X-ray absoptiometry predicted three- and four-component model body composition slightly less well than densitometry or deuterium dilution (both of which greatly influence these multi-component models).

4. The hydration fraction of fat-free mass was calculated to be 0.7382 ± 0.0213 (range 0.6941–0.7837) and the density of fat-free mass was 1.1015 ± 0.0073 kg/l (range 1.0795–1.1110 kg/l), with no significant difference between men and women for either.

5. The results suggest that the three- and four-component models are not compromised by errors arising from individual techniques. Dual-energy X-ray absorptiometry would appear to be a suitable alternative method for the assessment of body composition in these healthy adults. The traditional mean value assumed for density of the fat-free mass in classic densitometry (1.1 kg/l) appears to be appropriate, and the mean hydration fraction was close to values which are generally applied to the fat-free mass (0.72–0.73). Despite concealing considerable inter-individual variation, these mean values may be applied to groups with characteristics similar to those in this study. Finally, with the notable exception of skinfold thickness, bedside prediction methods show poor agreement with both the three- and the four-component models.

INTRODUCTION

Dual-energy X-ray absorptiometry (DEXA) is being increasingly used for the prediction of gross body composition because of its ability to differentiate between bone mineral, and fat and fat-free soft tissues [1]. Despite its relatively high cost, DEXA has certain advantages over other reference methods (densitometry and deuterium dilution) in view of its speed and ease of application to a variety of subjects.

Classical two-component models (fat and fat-free mass) for the measurement of body composition are limited by assumptions regarding the constancy of composition of fat-free mass. A three-component model (fat, water and fat-free dry matter [2]), which is based on measurements obtained from both densitometry and deuterium dilution, overcomes some uncertainties concerning the hydration fraction of fat-free mass, but it does assume a constant ratio of protein to mineral (the fat-free dry matter in this model). On theoretical grounds, a further improvement may be achieved by the use of a four-component model (fat, protein, water and mineral [3, 4]), which segregates protein from mineral. This model removes the need for some of the assumptions, inherent in the two- and three-component models, regarding proportions and average densities of different components of the fat-free mass. The four-component model incorporates direct measurement of total-body bone mineral content, as well

Key words: body fat, densitometry, dual-energy X-ray absorptiometry, fat-free mass, total-body water.

Abbreviations: BMI, body mass index; DEXA, dual-energy X-ray absorptiometry.

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as the established measurements of body density and total-body water. However, the densities of fat and protein are assumed to be constant, as are both the density of total-body mineral and the ratio of osseous to non-osseous mineral.

The aims of this study were four-fold: first, to establish the extent of agreement between the various methods of body composition analysis, with emphasis on DEXA and the three- and four-component models; secondly, to quantify the extent of the variation in density of the fat-free mass in this sample population, and to test whether differences in mean values exist between men and women; thirdly, to establish the hydration fraction of the fat-free mass and, once again, to evaluate any putative gender differences; and finally, to assess the extent to which measurement errors of the individual reference techniques are propagated in the determination of body composition by multi-component models.

METHODS

Subjects

Twenty-eight healthy adults (12 women and 16 men) volunteered to participate in the study. The characteristics of these subjects are shown in Table 1. Approval for the study was granted by the Ethical Committee of the Dunn Clinical Nutrition Centre, and all subjects gave their informed written consent.

Weight and height

Body weight, corrected to nude weight, was measured using electronic scales with a digital read-out (Sauter Type E1210; Todd Scales, Newmarket, Suffolk, U.K.). Height was measured with a wall-mounted stadiometer (Holtain Ltd, Crosswell, Crymych, Dyfed, U.K.).

DEXA

Total-body fat, fat-free soft tissue and bone mineral ash measurements were obtained for all subjects using a Lunar DPX whole-body scanner (Lunar Radiation Corporation, Madison, WI, U.S.A.) as described elsewhere [5] and were analysed using the Lunar DPX software package 3.1.

Densitometry

Body volume and density were assessed by the technique of Akers & Buskirk [6], modified by the use of a helium-dilution technique for the measurement of lung volume (based on that of Gnaedinger *et al.* [7]). In the calculation of body composition using this technique, the density of fat was assumed to be 0.9007 kg/l and the density of the fat-free tissue was assumed to be 1.100 kg/l [2].

Isotope-dilution technique

Total-body water volume (litres) was determined from measurement of isotope dilution, using saliva samples obtained at 4, 5 and 6 h after dosing [8]. Phantom dilutions were made at 20°C, and the corresponding volumes at 36°C were calculated from the densities of water at 20°C and 36°C (0.99823 kg/l and 0.99371 kg/l, respectively, Smithsonian Tables). Since the density of water at 20°C (0.99823 kg/l) is virtually identical with that found for deionized Cambridge tap water (0.9985 kg/l [8]), the difference was considered small enough to be ignored. In calculating total-body water, it was assumed that deuterium-dilution space was a factor of 1.04 times greater than total-body water, owing to proton exchange [9], and for calculating body composition it was assumed that the hydration fraction of the fat-free mass was 0.7194 (calculated from the study by Siri [2]).

Whole-body potassium measurement

Estimates of body fat and fat-free mass were obtained from whole-body ^{40}K measurements [9, 10].

Prediction or bedside methods

Skinfold thickness was measured at four sites with standard skinfold calipers (Holtain Ltd), and estimates of body fat were obtained by the method of Durnin & Womersley [11] using age and sex appropriate equations.

Table 1. Characteristics of subjects

	Women ($n=12$)		Men ($n=16$)		Combined ($n=28$)	
	Mean ± SD	Range	Mean ± SD	Range	Mean ± SD	Range
Age (years)	31.8 ± 11.0	20–59	33.8 ± 10.7	18–55	32.9 ± 10.6	18–59
Height (m)	1.65 ± 0.06	1.56–1.78	1.77 ± 0.06	1.64–1.88	1.72 ± 0.09	1.56–1.88
Weight (kg)	56.89 ± 6.25	48.25–67.80	73.71 ± 9.07	59.39–90.06	66.50 ± 11.55	48.25–90.06
BMI (kg/m^2)	20.86 ± 2.08	17.10–24.65	23.40 ± 2.19	19.84–28.10	22.31 ± 2.46	17.10–28.10
Fat-free mass (kg)*	42.10 ± 3.18	36.60–46.63	58.74 ± 7.39	49.28–77.83	51.61 ± 10.24	36.60–77.83
Fat (%)*	25.63 ± 4.96	19.56–37.95	19.76 ± 4.26	12.01–25.04	22.28 ± 5.38	12.01–37.95
Total-body water (litres)	31.38 ± 2.57	26.13–34.76	43.05 ± 5.40	36.94–57.62	38.06 ± 7.32	26.13–57.62

*Obtained from the four-component model.

Whole-body bioelectrical resistance/impedance was measured using the technique described previously [12]. These measurements were used to estimate total-body fat mass (as a percentage of body weight) and fat-free mass (kg) using equations (details unknown) supplied in the analyser software (Valhalla Scientific, San Diego, CA, U.S.A.). Estimates of body fat, fat-free mass and water were obtained from near-i.r. interactance measurements by the method described by Elia *et al.* [13]. Body mass index (BMI) was calculated from weight and height (kg/m^2), enabling estimation of body fat from previously published prediction equations [14].

Three-component model

For the purposes of this study, a three-component model of body composition was constructed on the basis of three major and distinct chemical components within the body: fat, water and the remaining fat-free dry mass (consisting of protein and mineral in constant ratio). Body composition was calculated by using combined measurements of body weight, total-body water (from deuterium dilution) and body volume (from densitometry).

This three-component model was based on the following assumptions [15]: that the density of body fat was a constant 0.9007 kg/l, that the density of water at 36°C was 0.99371 kg/l and that the density of protein plus mineral was 1.5157 kg/l [obtained when protein (density 1.34 kg/l) and mineral (density 3.0375 kg/l) were combined in the assumed whole-body ratio of 0.7926:0.2074].

The fraction of body fat (f) was calculated according to the following general equation, a rearrangement of that of Keys & Brozek [16]:

$$f = d_1(d_2 - D)/D(d_2 - d_1)$$

where, for the purposes of the three-component model, d_1 represents the density of fat, d_2 represents the density of protein combined with mineral (constant ratio in the whole body, as above), and D represents the density of fat combined with this same ratio of protein and mineral (D_{f+pm}).

With substitution of the assumed density values (see above) into the equation of Keys & Brozek [16]:

$$f = [2.21989/(D_{f+pm})] - 1.46462$$

The only unknown in this equation, D_{f+pm}, was calculated from the general equation: density = mass/volume, where mass and volume of fat plus protein/mineral were obtained by the difference between the whole-body and the total-body water component:

$$D_{f+pm} = (\text{body mass} - \text{water mass})/(\text{body volume} - \text{water volume})$$

In terms of the basic measurements:

$$\text{Fat (kg)} = 2.220\text{BV} - 0.764\text{TBW} - 1.465\text{Wt.}$$

where BV = body volume in litres (from densitometry), TBW = total-body water volume in litres (from deuterium dilution) and Wt. = body weight in kg.

Clearly, although only the equation for fat is shown, fat-free mass is also obtainable from this model.

Four-component model

The four-component model was based on principles similar to those adopted for the three-component model, but involved the additional measurement of bone mineral by DEXA, allowing the body to be segregated into fat, water, mineral and protein.

The density of total-body mineral (using data from Brozek *et al.* [15]) was calculated to be 3.0375 kg/l, assuming a constant ratio (0.8191:0.1809) of osseous (bone) mineral (density = 2.982 kg/l) to non-osseous mineral (density = 3.317 kg/l).

Total-body mineral mass was obtained from the DEXA estimate of bone ash:

$$\text{Total-body mineral mass} = \text{ash} \times 1.2741 \text{ kg}$$

where the fraction of osseous mineral comprising ash was assumed to be 0.9582, and, from the ratio of osseous to non-osseous mineral, the mass of non-osseous mineral was assumed to be ash × 0.23048 kg (calculated from Brozek *et al.* [15]).

Total-body mineral volume was obtained from total-body mineral mass and mean density of osseous and non-osseous mineral, thus:

$$\text{Total-body mineral volume} = \text{ash} \times (1.2741/3.0375) \text{ or ash} \times 0.41946 \text{ litres}$$

The fraction of body fat (f) was then calculated in a manner analogous to that used in the three-component model [in this case, d_2 of the equation of Keys & Brozek [16] (see above) represents protein alone, and D represents the combined density of fat and protein, D_{f+p}]. Hence

$$f = [2.7474/(D_{f+p})] - 2.0503$$

where the term D_{f+p} was calculated from the difference between whole-body mass and total-body water plus mineral mass divided by the difference between whole-body volume and total-body water plus mineral volume, in a manner analogous to the three-component model:

$$D_{f+p} = [\text{body mass} - (\text{water mass} + \text{mineral mass})]/[\text{body volume} - (\text{water volume} + \text{mineral volume})]$$

In terms of the basic measurements:

$$\text{Fat (kg)} = 2.747\text{BV} - 0.710\text{TBW} + 1.460A - 2.050\text{Wt.}$$

where the abbreviations BV, TBW and Wt. are the same as in the three-component model and A = ash in kg (from DEXA).

Protein (kg) may be calculated in a similar manner to fat, and fat-free mass is also available using this model.

Calculation of density and hydration of fat-free mass

The density of fat-free mass (D_{ffm}) was calculated from the mass and volume of each individual component (water, protein and mineral) using the four-component

model, thus:

$$D_{ffm} = (\text{mass of water} + \text{protein} + \text{mineral}) / (\text{volume of water} + \text{protein} + \text{mineral})$$

In terms of the basic measurements:

$$D_{ffm} = (0.710\text{TBW} + 3.050\text{Wt.} - 2.747\text{BV} - 1.460A) / (0.788\text{TBW} + 2.276\text{Wt.} - 2.050\text{BV} - 1.621A)$$

where the abbreviations TBW, Wt., Bv and A are as defined for the four-component model.

The hydration fraction was calculated from the mass of total-body water (from deuterium dilution) and total-body fat-free mass, obtained from the four-component model.

In terms of the basic measurements:

$$\text{Hydration fraction} = \text{TBW}/(0.715\text{TBW} + 3.070\text{Wt.} - 2.765\text{BV} - 1.469A)$$

where the abbreviations TBW, Wt., BV and A are as defined for the four-component model.

Statistics

The bias and 95% (± 2 SD) limits of agreement [17] for comparison of the estimates of body composition by a particular method compared with other methods were established as previously in our laboratory (e.g. Fuller & Elia [12]). Student's t-test was used to test for the significance of any differences between population samples studied here (male versus female).

Estimates of the overall measurement precision in terms of fat or fat-free mass were calculated for each model of body composition from analysis of the propagation of errors. These estimates were based on a 70 kg man consisting of 15% fat, a hydration fraction of fat-free mass of 0.7194 (total-body water volume is therefore 42.8 litres) and a total-body bone mineral ash content of 3.2 kg. The values adopted for errors in the basic measurements were as follows: body density, 0.0025 kg/l [2]; body weight, 0.01 kg [18]; body volume (derived from body weight and density), 0.157 litres; total-body water volume, 0.45 litres; total-body bone mineral ash, 0.03 kg [5]. The precision for estimates of total-body water was based on sequential measurements of the isotopic enrichment of water in saliva samples taken at 4, 5 and 6 h after oral administration of the isotope. The results for all subjects were compared with the enrichment of a single phantom dilution of the stock dose. Precision for the measurement of water calculated from this study was 0.45 litres (about 1%), although a precision of 0.86 litres (2%), which may be more appropriate under certain circumstances [18], was also considered.

RESULTS

The bias and 95% limits of agreement (shown as bias ± 2 SD) between methods are given in Table 2 for percentage of body weight as fat, and in Table 3 for fat-free mass (kg). In many of the comparisons, such as the three- versus the four-component model, there is no material difference in the bias and 95% limits of agreement between men and women. However, in others, for instance densitometry versus the BMI formula, the bias is seen to be of opposite sign and the 95% limits of agreement differ in magnitude between men and women. Therefore, results obtained for each gender separately have been deposited as *Clinical Science* Tables A and B 92/2 with the Librarian, Royal Society of Medicine, 1 Wimpole Street, London W1M 8AE, U.K., from whom copies are available on request. Table 4 shows the results of this same statistical analysis performed on the alternative methods alone (for all subjects combined). Clearly, there is less overall bias and better agreement when reference methods are compared with each other than when the alternative methods are compared (either with reference methods or with each other).

The density of the fat-free mass (mean $\pm$ SD) was found to be 1.1003 ± 0.0066 kg/l and 1.1024 ± 0.0078 kg/l for women and men respectively, with no significant difference between the sexes. The overall mean was thus 1.1015 ± 0.0073 kg/l with a range of 1.0795–1.1110 kg/l.

Table 2. Comparison of various body composition techniques with some reference methods: bias and 95% (± 2SD) limits of agreement for estimates of body fat (as percentage of body weight). The Table shows the reference method/model (top row of the Table) minus the alternative method (left-hand column of the Table). Values for the bias for fat-free mass as a percentage of body weight are equal and opposite to those for percentage fat, and the 95% limits of agreement (bias ± 2 SD) are equal for both (see [17]).

	Four-component model	Three-component model	DEXA	Densitometry	Deuterium dilution
Three-component model	1.23 ± 1.43	—			
DEXA	1.36 ± 4.95	0.13 ± 5.60	—		
Densitometry	0.75 ± 3.78	−0.47 ± 4.13	−0.60 ± 4.46	—	
Deuterium dilution	1.70 ± 3.91	0.48 ± 3.15	0.34 ± 7.50	0.95 ± 6.83	—
BMI	0.71 ± 8.21	−0.51 ± 8.46	−0.65 ± 8.68	−0.04 ± 9.09	−0.99 ± 8.71
Skinfold thickness	0.09 ± 5.42	−1.14 ± 5.57	−1.27 ± 5.42	−0.66 ± 5.00	−1.61 ± 7.13
Whole-body impedance	3.39 ± 7.30	2.16 ± 7.87	2.03 ± 7.07	2.63 ± 7.87	1.69 ± 8.62
Whole-body potassium	−0.60 ± 10.42	−1.83 ± 10.78	−1.96 ± 8.66	−1.35 ± 10.23	−2.30 ± 12.40
Near i.r. interactance	0.41 ± 7.79	−0.81 ± 8.60	−0.94 ± 6.12	−0.34 ± 7.49	−1.29 ± 10.15

Table 3. Comparison of various body composition techniques with some reference methods: bias and 95% (±2 SD) limits of agreement for estimates of fat-free mass (in kg). The Table shows the reference method/model (top row of the Table) minus the alternative method (left-hand column of the Table). Values for the bias for kg of fat are equal and opposite to those for fat-free mass, and the 95% limits of agreement (bias ± 2 SD) are equal in both (see [17]).

	Four-component model	Three-component model	DEXA	Densitometry	Deuterium dilution
Three-component model	−0.74 ± 1.02	—			
DEXA	−1.16 ± 4.07	−0.42 ± 4.48	—		
Densitometry	−0.69 ± 3.19	0.06 ± 3.57	0.48 ± 2.98	—	
Deuterium dilution	−1.37 ± 2.97	−0.63 ± 2.57	−0.20 ± 5.31	−0.68 ± 4.65	—
BMI	−0.54 ± 6.34	0.20 ± 6.54	0.63 ± 5.91	0.15 ± 6.05	0.83 ± 6.46
Skinfold thickness	−0.20 ± 3.88	0.54 ± 4.04	0.96 ± 3.43	0.48 ± 3.21	1.16 ± 5.03
Whole-body impedance	−2.43 ± 6.04	−1.69 ± 6.43	1.27 ± 4.75	−1.75 ± 5.29	−1.06 ± 6.30
Whole-body potassium	0.05 ± 8.17	0.80 ± 8.48	1.22 ± 6.04	0.74 ± 7.16	1.42 ± 8.90
Near i.r. interactance	−0.71 ± 5.84	0.03 ± 6.39	0.45 ± 3.87	−0.29 ± 4.88	0.65 ± 6.91

Table 4. Comparison of various alternative body composition techniques: bias and 95% (±2SD) limits of agreement for estimates of fat and fat-free mass. The Table shows the reference method/model (top row of the Table) minus the alternative method (left-hand column of the Table). Values for the bias for fat-free mass as per cent body weight and those for fat mass (kg) are equal and opposite to those for per cent fat and fat-free mass (kg), respectively, and the 95% limits of agreement (bias ± 2SD) are equal for both (see [17]).

	BMI	Skinfold thickness	Whole-body impedance	Whole-body potassium
Fat (% of body wt.)				
Skinfold thickness	−0.62 ± 7.77	—		
Whole-body impedance	2.68 ± 5.11	3.30 ± 7.08	—	
Whole-body potassium	−1.31 ± 14.65	−0.69 ± 11.38	−4.01 ± 12.82	—
Near-i.r. interactance	−0.30 ± 9.70	0.33 ± 8.61	−2.97 ± 7.98	−1.01 ± 11.07
Fat-free mass (kg)				
Skinfold thickness	0.34 ± 5.14	—		
Whole-body impedance	−1.89 ± 3.85	−2.23 ± 4.86	—	
Whole-body potassium	0.59 ± 10.08	0.26 ± 7.63	2.49 ± 8.71	—
Near-i.r. interactance	−0.18 ± 6.68	−0.51 ± 5.61	1.72 ± 5.31	−0.77 ± 7.25

The hydration fraction of the fat-free tissue (mean ± SD) was calculated to be 0.7449 ± 0.0192 for women and 0.7332 ± 0.0219 for men, with no significant difference between them. Thus the mean hydration fraction of all subjects combined was calculated to be 0.7382 ± 0.0213 with a range of 0.6941–0.7837.

Precision (SD) associated with estimated amounts of fat (kg) or fat-free mass (kg) for a 70 kg man using the three- and four-component models was found to be ± 0.49 kg and ± 0.54 kg, respectively, when a 1% precision for water estimation was assumed. Alternatively, assuming a 2% precision for water estimation (other measurement precisions being held constant), the corresponding values were ± 0.74 kg and ± 0.75 kg, respectively. The precision found for densitometry was ± 0.78 kg and that for deuterium dilution was ± 0.62 kg, assuming 1% precision, and 1.20 kg when 2% precision was considered. The precision associated with the calculation of hydration fraction was calculated to be ± 0.0066, and that for the density of fat-free mass was ± 0.0020 kg/l (only the 1% precision in water estimate is shown here because the errors from this source largely cancelled out, see the equations for calculating density and hydration fraction of fat-free mass). The largest contribution to the imprecision associated with densitometry originated from the estimate of body volume (± 0.77 kg). Precision in the three- and four-component models, and in density and hydration fraction of fat-free mass, was found to be mostly attributable to the estimation of total-body water and body volume, with the contribution of body weight and mineral ash being negligible.

Substitution of the assumed and commonly accepted values for either density (1.1 kg/l) or hydration fraction (0.72–0.73) of the fat-free mass by values representing the extremes of the range found in this study demonstrated potential errors in the estimation of fat (and fat-free mass) of up to 2.4 kg and 2.6 kg, respectively. These represent about a 25% error in the fat estimation (and about 5% in fat-free mass) of a 70 kg reference man.

DISCUSSION

This study has highlighted the potential use of DEXA for assessing body composition. It is simple to perform

and exposes the volunteer or patient to only a low level of radiation [5]. Determination of fat (percentage of body weight) or fat-free mass (kg) by DEXA agreed slightly less well with the three- or four-component models than did densitometry or deuterium dilution. This is not very surprising, since densitometry and deuterium dilution provide the only measured determinants of body fat in the three-component model, and are the major determinants of fat in the four-component model. Moreover, DEXA was found to be the best predictor of mean body composition obtained by four separate two-component reference methods (densitometry, deuterium dilution, DEXA and total-body potassium, results not shown). Although, of these two-component methods, total-body potassium predicted body composition with least accuracy and agreement (compared with either the reference methods/models or the mean of the two-component methods), this does not necessarily imply that all models based on whole-body potassium have this predisposition. The whole-body counter used in this study had only two sodium iodide crystals, which yield relatively poor counting precision (coefficient of variation greater than 5% [19]).

The application of bedside prediction methods to estimate body composition has certain advantages over the more sophisticated primary methods in terms of simplicity, convenience and relative cost. Irrespective of which reference method is selected for comparison, the use of the sum of four skinfold thicknesses [11], measured by a single observer, has proved to be a better predictor of body composition than other bedside prediction methods (equations based on BMI, bioelectrical impedance/resistance and near-i.r. interactance) in this group of subjects.

Certain components of the body were not taken into consideration in the three- and four-component models. Of these, nucleic acids and glycogen probably represent the greatest fractions. However, their mass in the whole body is relatively low (approximately, 0.7 kg and 0.5 kg, respectively, corresponding to about 1% and less than 1% body weight) and their densities (DNA, 1.42 kg/l; RNA, 1.49 kg/l; glycogen, 1.52 kg/l) are not so very different from that of protein (1.34 kg/l). Hence, the errors involved as a result of neglecting nucleic acids and glycogen from these models are considered to be small, and, for the most part, these components are likely to be included with the protein fraction [4]. Similarly, amino acids (a 70 kg reference man contains about 250 g of free amino acids, the densities of which are both above and below 1.34 kg/l depending on the particular amino acid), urea (10–15 g in a 70 kg man, density 1.32 kg/l) and other minor components, such as uric acid and creatinine, are partitioned mostly with the protein fraction; this is because the densities of these components are closer to the density of protein (1.34 kg/l) than to that of fat (0.9 kg/l).

The four-component model also provides important information concerning the likely variation in density of the fat-free mass. In classical densitometry this value is generally assumed to be 1.1 kg/l. Deviations from this value will result in errors in the prediction of body composition. The calculations in this study suggest that there is considerable inter-individual variation in the density of fat-free mass, despite the close proximity of the mean value to 1.1 kg/l for both men and women. Since these differences do exist, both within and between populations, a single value for the density of the fat-free mass should not be universally applied. For example, either osteoporosis or overhydration (oedema) would tend to decrease the mean density of fat-free mass, whereas greater proportions of bone mineral (as found in some Negroid populations [20]) would tend to increase it.

The four-component model of body composition also enables the hydration fraction of fat-free mass to be assessed. In water-dilution techniques, the value for hydration fraction of the fat-free mass applied to body composition assessment ranges from 0.7194 to 0.7320. The results of this study suggest that it is the latter value which is the more appropriate for groups of individuals with similar characteristics to those studied here (mean hydration fraction calculated to be 0.7382, with no significant difference between men and women). Furthermore, applying the former value (0.7194, as suggested by calculation from the data of Siri [2]) to the deuterium-dilution technique (in isolation) of body composition assessment is not appropriate in our group of subjects because it overestimates fat-free mass relative to the four-component model (for both men and women). However, despite the use of the most appropriate mean hydration fraction for the water-dilution techniques (two-component model), there may still be substantial error in body composition analysis because of inter-individual variation. Perhaps surprisingly, the hydration fractions of the fat-free mass assumed for reference man and reference woman [21] differ substantially (0.74 and 0.69, respectively). To establish these values, the Task Group on Reference Man [21] considered measurements of water obtained from one group of individuals and measurements of fat-free mass from another. It is suggested that this approach is inappropriate, and may have been responsible for the apparent discrepancy. By the use of the more consistent experimental approach adopted in this study, an appropriate value for the hydration fraction of both men and women has been identified.

Although many of the above difficulties may be overcome by the use of the four-component model, there is some concern that measurement errors arising from the individual techniques may be propagated to produce a large error in the final estimate of fat and fat-free mass. However, the present analysis suggests that the individual measurement errors are not substantially additive, and that the overall measurement errors for both fat and fat-free mass amount to only about 1% or less of body weight (which is largely due to the measurement of body water and body volume). This error is as low or lower than measurement errors associated with the determination of body fat using either densitometry or deuterium-dilution in isolation.

The precision (SD) for the estimate of the hydration fraction (± 0.0066) and the density (± 0.0020 kg/l) of fat-

free mass was found to correspond to approximately one-third of the inter-individual variation (± 0.02 and ± 0.0073 kg/l, respectively). This variability, which cannot be accounted for by propagation of measurement error, is considered to be largely due to biological variation in the relative proportions of components of the fat-free mass, but it may also be due to the use of incorrect assumptions in the four-component model. These assumptions relate to components which remain unaccounted for by the model (discussed above), the densities of various components which are considered in the model, the use of a factor for total-body water which is 1.04 times greater than the measured deuterium-dilution space, and the whole-body ratio of osseous relative to non-osseous mineral. Of these assumptions, the greatest level of confidence is associated with the assumed densities of water and fat. In contrast, there is relatively less confidence in the ubiquitous application of the remaining assumptions. For example, since body proteins comprise a heterogeneous group of a large number of different proteins with different densities, the proportions in which they are to be found in the whole body may vary between different individuals or in different circumstances. However, mammalian muscle proteins, which are the most abundant proteins in the body, have a density estimated to be between 1.33 and 1.35 kg/l (calculated from data taken from Mendez & Keys [22]). Collagen, which is estimated to account for 25–30% of total protein in the human body (mainly in bone and skin), is considered to have a density of 1.36 kg/l [23]. These values are close to the value for the density of protein applied to the models used in this study.

It is concluded that DEXA provides a suitable alternative method to densitometry or deuterium dilution for the assessment of body composition. Furthermore, skinfold thickness measurements predicted body composition (as assessed by the reference methods) better than any of the other bedside methods. In addition, values are proffered (derived from the four-component model) which represent the mean density and hydration fraction of the fat-free mass in healthy non-obese adults with similar characteristics to those in this study.

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