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Original article

Prediction of body composition in anorexia nervosa: Results from a retrospective study

Maurizio Marra [a], Rosa Sammarco [a, *], Emilia De Filippo [a], Annarita Caldara [a], Enza Speranza [a], Luca Scalfi [b], Franco Contaldo [a, c], Fabrizio Pasanisi [a, c]

[a] Department of Clinical Medicine and Surgery, University Federico II, Naples, Italy
[b] Department of Public Health, Federico II University of Naples, Italy
[c] Inter-University Centre for Obesity and Eating Disorders (CISRODCA), Federico II University of Naples, Italy

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SUMMARY

Background & aims: The assessment of body composition is crucial in evaluating nutritional status in female subjects with anorexia nervosa (AN) and improving their clinical management. The aim of this retrospective study was to assess the accuracy of selected BIA (bioimpedance analysis) equations for fat-free mass (FFM) in female AN subjects and to formulate a specific equation for these subjects.
Methods: Eighty-two restrictive female AN subjects (age 20.5 ± 3.7 yrs, BMI 15.7 ± 1.7 kg/m^2) were studied. Body composition was determined with dual-energy X-ray absorptiometry (DXA) and estimated by BIA using five different equations. Linear correlation analysis was carried out to evaluate the association of FFM with selected variables. Multiple regression analysis was used to formulate specific equations to predict FFM in AN.
Results: All predictive equations underestimated FFM at the population level with a bias from −5.6 to −11.7%, while the percentage of accurate predictions varied from 12.2% to 35.4%. More interestingly, multiple regression analysis clearly indicates that, in addition to weight, ZI_{100} or RI also emerged as independent predictors of DXA-derived FFM, increasing the prediction power of the equation well above that observed with anthropometric characteristics only.
Conclusions: This study shows that the selected predictive BIA equations considered exhibit an insufficient accuracy at the population and the individual level. Predictive formulas based on body weight plus BIA parameters such as RI and ZI_{100} offer a rather accurate prediction of FFM (with high R squared).

1. Introduction

Anorexia nervosa (AN), which is a psychiatric disorder characterized by both underweight and intense fear of gaining weight or becoming fat [1], is a form of protein-energy malnutrition due to decreased food intake and/or compensatory actions such as vomiting, laxative abuse and increased physical activity [2,3].

Body weight and body mass index (BMI, kg/m^2) are widely used to establish the degree of underweight. However, when malnutrition is severe, body weight is not a reliable method for determining nutritional status [4,5]; for instance, lost body cell mass can be replaced by extracellular fluid [4–6]. Thus, evaluating body composition is crucial for nutritional assessment in AN subjects and improving their clinical management. For this purpose, different non-invasive techniques may be used, such as dual-energy X-ray Absorptiometry (DXA), often considered as a reference method, bioelectrical impedance analysis (BIA) and skinfold thickness measurement.

There is evidence in the literature [7,8] that in young adults DXA shows high levels of accuracy in estimating body composition compared with a four-component model [9]. More specifically, a strong correlation between DXA and Computed Tomography was recently found in adult AN individuals irrespective of the level of hydration [10]. Actually, DXA is not so far routinely used in the clinical assessment of such subjects, as is the case with BIA [11,12]. Few studies have examined the limitations of BIA in AN [11], and there are currently no widely accepted disease-specific equations for estimating body composition in these subjects [12,13]. Previously, Scalfi et al. [14] predicted total body water (TBW) and Bedogni et al. [15] fat-free mass (FFM) with BIA in small groups of AN

* Corresponding author. Department of Clinical Medicine and Surgery, Federico II University Hospital, Via S. Pansini 5, 80131, Naples, Italy. Fax: +39 0815466152.
E-mail address: rosa.sammarco@unina.it (R. Sammarco).

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and control women, using deuterium oxide (D_2O) and DXA as reference method, respectively. More recently, Mattar et al. [16] studied underweight AN subjects, comparing measurements of FFM and fat mass (FM) using DXA and five different BIA equations already validated in healthy populations, showing large differences in accuracy.

Based on this background, the aim of our study was to evaluate the accuracy of selected BIA equations used to estimate body composition in AN and to identify significant predictors in order to propose new predictive equations of FFM in AN female subjects.

2. Materials and methods

2.1. Individuals

A group of 82 clinically stable restrictive anorexia nervosa patients (DSM-IV, 1994) attending the outpatient clinic of the Clinical Nutrition Unit, Department of Clinical Medicine and Surgery, Federico II University Hospital, Naples, Italy, was considered in this retrospective study (between October 2009 and December 2014).

All patients underwent routine laboratory, clinical evaluations, BIA and DXA in order to support the reliability of widely used predictive equations for body composition and to promote their use in clinical practice. Subjects or parents, when required, gave informed consent for routine diagnostic evaluations. All measurements were performed with a standardized protocol and carried out by experienced staff, in fasting conditions and early in the morning.

2.2. Anthropometry

Body weight was measured to the nearest 0.1 kg with a platform beam scale and height to the nearest 0.5 cm with a stadiometer (Seca 709 and SECA 220, respectively; Seca, Hamburg, Germany). BMI was calculated as body weight (kg) divided by height squared (cm^2).

2.3. DXA

DXA was performed on the whole body using an Prodigy Primo Lunar, A223040501, General Electric Company, Madison (v13.31 software). No special preparation was required; all participants had their underwear on (no metal accessories worn during measurement). DXA uses an X-ray generating source, with two X-ray beams with different energy levels. FFM (lean mass + bone mineral content) and FM are assessed based on their X-ray attenuation properties.

2.4. Bioelectrical impedance analysis

BIA was performed on the non-dominant side of the body in standard conditions: postabsorptive state, at a room temperature between 22 and 25 °C, with the subject being in the supine position for 10 min after voiding [17]. In single-frequency BIA resistance (R, ohm), reactance (Xc, ohm) and phase angle (PhA, degrees) were measured at 50 kHz (BIA 101 analyzer, Akern, Florence), while in multifrequency BIA impedance (Z) was determined at 5, 50 and 100 kHz (Z_5, Z_{50} and Z_{100}) with a Human Im Plus II analyzer (DS Medica, Milan). Subsequently, resistance index (RI = $height^2/R$, cm^2/ohm) and Z indexes at 5, 50, 100 kHz (ZI = $height^2/Z$, ZI_5, ZI_{50} and ZI_{100}, cm^2/ohm) were calculated. For the measurement, four disposable electrodes were placed as follows: a) two injecting electrodes on the dorsum of hand, proximal to the metacarpal-phalangeal joint line, and on the dorsum of foot, proximal to the metatarsal-phalangeal joint line; b) two sensing electrodes on the mid dorsum of wrist, on the line joining the bony prominences of radius and ulna, and ankle, on a line joining the bony prominences of the medial and lateral malleoli. Both the instruments were regularly checked using resistors and capacitors of known values. BIA was also measured in six individuals on subsequent days: all the differences were within ±2% for R, Xc, and Z at different frequencies, and ±4% for PhA.

Three general BIA equations for predicting FFM were chosen according to appropriate validation in healthy Caucasian subjects and applicable age range.

Deuremberg et al. [18] for females:

$$FFM = -12.44 + 0.34 \times RI + 0.1534 \times height + 0.273 \times weight - 0.127 \times age$$

Kyle et al. [19] for females:

$$FFM = -4.104 + 0.518 \times RI + 0.231 \times weight + 0.130 \times Xc$$

Sun et al. [20] for females:

$$FFM = -9.529 + 0.696 \times RI + 0.168 \times weight + 0.016 \times R$$

The specific equations used to evaluate body composition in anorexia nervosa were:

Scalfi-1 [14]:

$$TBW^* = 0.434 \times weight + 6.326$$

Scalfi-2 [14]:

$$TBW^* = (0.563 \times ZI_{100} + 2.695)$$

Bedogni [15]:

$$FFM = 0.6 \times ZI_{50} + 0.2 \times weight + 3.3$$

(*TBW was converted into FFM assuming a 73% water in FFM).

2.5. Statistical analysis

Results are expressed as mean and standard deviation plus minimum and maximum value.

The accuracy of the predictive equations was calculated both at population and individual level. The mean bias was considered as a measure of accuracy at a population level [21]; a reliable estimate was defined as a mean percentage difference ±5% between BIA and DXA-derived FFM. On the other hand, the percentage of individuals with a BIA-DXA difference within ±5% was considered as a measure of accuracy at an individual level. Values lower than 95% indicated underprediction and values higher than 105% overprediction. Finally, the root mean squared prediction error (RMSE) was used to better assess predicted-measured differences in absolute values (no sign).

Linear correlation analysis was carried out to evaluate the association between body composition (FFM and FM) and selected variables (see below), while multiple regression was used to derive equations to predict FFM in female subjects with anorexia nervosa. In both cases, three different sets of variables were considered:

1) general characteristic of individuals: age, height, weight and BMI;
2) single-frequency BIA: R, Xc, RI and PhA;
3) multifrequency BIA (kHz): Z_5, Z_{50}, Z_{100}, ZI_5, ZI_{50} and ZI_{100}.

The significant level was set at $p < 0.05$. All statistical analysis was performed using SPSS (vers. 20.0).

3. Results

Table 1 shows anthropometric data, BIA variables and DXA-derived measures of body composition in the 82 restrictive AN female subjects studied (mean age 20.5±3.7 yrs, height 159.0±6.7 cm, weight 40.0±5.3 kg, BMI 15.7±1.7 kg/m^2).

3.1. Agreement between DXA and BIA in assessing FFM

Data on the agreement for FFM between DXA and BIA predictive equations are summarized in Table 2. All the predictive equations on average significantly underestimated DXA-derived FFM. The Sun equation exhibited the best agreement with a mean difference of −2.2 kg (mean bias −5.6%) while the Kyle equation performed the worst with a mean difference of −4.1 kg (mean bias −11.2%). RMSE ranged from 2.6 kg (Sun) to 4.3 kg (Kyle) (Table 2). The percentage of accurate predictions varied from 12.2% (Kyle) to 35.4% (Sun), that of underprediction from 5.6% (Scalfi-2) to 85.4% (Deurenberg), and that of overprediction from 0% (Deurenberg) to 79.6% (Scalfi-2).

3.2. Linear correlation between DXA-derived body composition and other variables

All the selected variables significantly correlated with FFM except age and Xc. The closer correlation was observed for weight ($r = 0.893$) in set 1, RI ($r = 0.725$) in set 2, and ZI_{100} ($r = 0.738$) in set 3 (Table 3). The association between FFM and ZI increased at higher frequencies, as suggested by a greater correlation coefficient (ZI_5 $r = 0.672$, ZI_{50} $r = 0.729$, ZI_{100} $r = 0.738$).

DXA-derived FM was significantly correlated with weight ($r = 0.657$) and BMI ($r = 0.565$), while in single-frequency BIA the most closely correlated parameters were PhA ($r = 0.400$) and Xc ($r = 0.301$). No significant correlation emerged with multifrequency BIA variables (Table 3).

Table 1
Anthropometric data, BIA parameters and body composition evaluated by DXA in 82 female subjects with anorexia nervosa.

		Mean	SD	Min	Max
Individual characteristics					
Age	yrs	20.5	4.7	16	30
Weight	kg	40.0	5.3	28.0	51.0
Height	cm	159.0	6.7	142	178
BMI	kg/m^2	15.7	1.7	10.3	18.4
Single-frequency BIA					
Resistance	ohm	715	111	487	1289
Reactance	ohm	61.3	16.5	25	117
Phase angle	degrees	4.89	0.98	2.29	6.83
RI	cm^2/ohm	36.1	5.4	21.1	50.5
Multifrequency BIA					
Z at 5 kHz	ohm	824	101	649	1174
Z at 50 kHz	ohm	737	88	591	1008
Z at 100 kHz	ohm	695	83	563	925
ZI at 5 kHz	cm^2/ohm	31.4	4.1	23.7	39.7
ZI at 50 kHz	cm^2/ohm	35.2	4.5	26.1	44.7
ZI at 100 kHz	cm^2/ohm	37.3	4.9	27.2	47.8
DXA-derived body composition					
Fat-free mass	kg	35.9	4.2	26.9	43.7
Fat mass	kg	4.0	2.3	1.0	9.0
Fat mass	%	9.8	5.0	3.5	21.1

RI: resistance index (height^2/resistance), Z: impedance, ZI: impedance index (height^2/impedance).

3.3. Multiple regression models to predict FFM

To examine the relationships between DXA-derived measures of body composition and predictors, a multiple regression analysis was carried out with FFM as dependent variable and potential predictive variables grouped together according to the aforementioned three sets. Table 4 shows the predictors included in the equations with highest R squared and lowest standard error of estimate (SEE):

1) weight and height ($R^2 = 0.808$; SEE = 1.86 kg), when only the general characteristics of individuals were considered;
2) weight and RI ($R^2 = 0.857$; SEE = 1.60 kg), for characteristics of individuals plus single-frequency BIA;
3) weight and ZI_{100} ($R^2 = 0.884$; SEE = 1.49 kg), for characteristics of individuals plus multifrequency BIA.

4. Discussion

This study has assessed the reliability of predictive BIA equations for FFM in restrictive female AN subjects, using DXA as reference method. None of the equations considered seems adequate to estimate body composition at an individual level. On the other hand, weight and ZI_{100} emerged both as simultaneous and valuable predictors of FFM in the selected subjects.

The assessment of body composition (FFM and FM) plays a key role in evaluating nutritional status in AN, either before or during nutritional rehabilitation [26] In the clinical setting BIA is a field method which is used for estimating TBW and FFM in different diseases, and also in AN subjects [14–16,26] by using predictive equations including raw BIA variables such as R, Z or PhA. Actually, disease-specific BIA equations are expected to be more reliable and effective in conditions, like anorexia nervosa, when marked changes in body composition occur.

On the average, all the BIA equations significantly underestimated DXA-derived FFM, with the Sun equation exhibiting the best agreement and the Kyle equation the worst. The mean difference was always >5%, indicating an insufficient degree of accuracy on a population basis. No direct comparison can be made with the paper of Mattar et al. [16], which also examined the use of BIA in AN, because of differences in subjects' age, BIA predictive equations chosen, statistical analysis of data, etc. Indeed, that paper also showed a relatively small mean difference in FFM between BIA estimates and DXA.

On an individual basis, the percentage of predicted values that were considered as accurate resulted to be quite low (from 12.2% to 35.4% depending on the equation, Table 2). For the BIA equations derived in healthy subjects [18–20] this might be due to the changes in body water distribution observed in AN. Indeed, similar findings also emerged for the disease-specific BIA equations selected [14–15]. Both these equations were derived from small samples of individuals. Furthermore, with respect to the Scalfi-2 equation, FFM was calculated assuming a normal body hydration (about 73%), which may not be the case in AN.

Overall, these findings support the goal of reassessing in a larger sample of individuals the association between FFM (from a reference method) and a number of potential predictive variables, in order to propose new disease-specific equations to predict FFM in AN female subjects.

The results of our study (Table 3) show that in simple correlation analysis weight is the variable that is the most strongly correlated with DXA-derived FFM, while ZI_{100} is the most powerful predictor among BIA variables. The association between FFM and ZI increased at higher frequencies, as suggested by a greater correlation coefficient, in line with the hypothesis that Z at higher frequencies are

Table 2
Evaluation of fat-free mass with different predictive equations in 82 female subjects with anorexia nervosa based on predicted-measured differences, Bias, Root Mean Square Error (RMSE) and percentage of accuracy.

BIA predictive equation	BIA-derived FFM (kg)	BIA-DXA difference (kg)	Bias %	RMSE	Accurate prediction %	Under-prediction%	Over-prediction%
General population equations							
Deurenberg	32.5 ± 3.6	−3.4 ± 1.9	−9.2	3.4	20.7	79.3	0
Kyle	31.8 ± 3.5	−4.1 ± 2.3	−11.2	4.3	12.2	85.4	2.4
Sun	33.7 ± 3.2	−2.2 ± 2.3	−5.6	2.6	35.4	57.3	7.3
Disease-specific equations for anorexia nervosa							
Scalfi-1	32.5 ± 3.1	−3.4 ± 2.0	−9.2	3.5	17.1	82.9	0
Scalfi-2	32.4 ± 3.7	−3.7 ± 2.9	−11.7	4.1	14.8	5.6	79.6
Bedogni	32.9 ± 3.9	−2.9 ± 2.4	−8.1	3.2	25.6	70.7	3.7

Bias = mean percentage difference between BIA-derived FFM and DXA-derived FFM
Accurate prediction = percentage of BIA-derived values within ±5% of DXA-derived FFM
Underprediction = percentage of BIA-derived values < 5% of DXA-derived FFM
Overprediction = percentage of BIA-derived values > 5% of DXA-derived FFM
All BIA-DXA difference are significantly ($p < 0.001$) different from zero.

Table 3
Linear correlation between DXA-derived values of fat-free mass (FFM) and fat mass (FM) and individual characteristics or BIA variables.

	FFM		FM	
	r	p<	r	p<
Individual characteristics				
Age	−0.038	NS	−0.006	NS
Weight	0.893	0.001	0.657	0.001
Height	0.574	0.001	0.211	0.10
BMI	0.593	0.001	0.565	0.001
BIA single-frequency				
Resistance	−0.447	0.001	0.016	NS
Reactance	−0.088	NS	0.301	0.01
Resistance index	0.725	0.001	0.047	NS
Phase angle	0.251	0.05	0.400	0.001
BIA multifrequency				
Impedance at 5 kHz	−0.340	0.05	0.079	NS
Impedance at 50 kHz	−0.399	0.01	0.047	NS
Impedance at 100 kHz	−0.411	0.01	0.034	NS
ZI at 5 kHz	0.672	0.001	0.025	NS
ZI at 100 kHz	0.729	0.001	0.070	NS
ZI at 250 kHz	0.738	0.001	0.086	NS

Table 4
Predictive equations of fat-free mass (FFM) based on different combinations of predictive variables.

Variable sets	Predictive equation: FFM (kg)	R^2	SEE (kg)
1	$0.660 \times$ weight $+ 0.079 \times$ height $- 3.04$	0.808	1.86
2	$0.705 \times RI + 0.092 \times X_C + 4.81$	0.624	2.61
3	$0.019 \times ZI_5 + 0.927 \times ZI_{100} - 14.10$	0.640	2.62
1 + 2	$0.571 \times$ weight $+ 0.237 \times RI + 4.49$	0.857	1.60
1 + 3	$0.575 \times$ weight $+ 0.271 \times ZI_{100} + 2.61$	0.884	1.49
1 + 2+3	$0.575 \times$ weight $+ 0.271 \times ZI_{100} + 2.61$	0.884	1.49

Variable set 1 = individual characteristics: age, weight, height, and BMI.
Variable set 2 = single-frequency BIA: resistance (R), reactance (X_C), resistance index (RI), phase angle.
Variable set 3 = multifrequency BIA: $Z_{(100,\ 50,\ 5)}$, $ZI_{(100,\ 50,\ 5)}$.
FFM determined by using DXA.
SEE = Standard error of estimate.

more strongly correlated with TBW and FFM. More interestingly, multiple regression analysis clearly indicates that, in addition to weight, ZI_{100} or RI emerged as simultaneous predictors of DXA-derived FFM, increasing the prediction power of the equation well above that observed with anthropometric characteristics only. Actually, in the female AN subjects studied the best predictive equation for FFM, as judged from R squared and SEE, includes both body weight and ZI_{100} ($R^2 = 0.884$; SEE = 1.49 kg). Thus, performing BIA at frequencies >50 kHz may be useful in assessing body composition because allowing a more appropriate evaluation of intracellular water [22]. This may be particularly relevant in underweight patients where changes in the ratio between intracellular water and extracellular water are expected [23–25].

In conclusion, the predictive BIA equations here selected exhibit an insufficient accuracy at a population and an individual level. As main finding, predictive formulas based on body weight plus BIA variables such as RI and ZI_{100} offer a more accurate prediction of FFM, with high R squared, than those based solely on weight and height. The relatively small sample of AN female subjects we studied represents a limitation of the study, suggesting that a more extensive evaluation in a larger group with a multicenter protocol is needed to confirm the finding of the present study.

Statement of authorship

F.P., M.M. designed research; A.C., E.D.F., R.S., E.S. conducted research; M.M., L.S. analyzed and discussed all data; M.M., F.C., R.S. wrote the paper. M.M. and F.P. had primary responsibility for final content. All authors read and approved the final manuscript.

Conflict of interest

None of the authors had any conflict of interest.

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