European Journal of Clinical Nutrition (2005) 59, 155–160

0954-3007/05 $30.00
www.nature.com/ejcn

ORIGINAL COMMUNICATION

Body water distribution in severe obesity and its assessment from eight-polar bioelectrical impedance analysis

A Sartorio[1], M Malavolti[2], F Agosti[1], PG Marinone[1], O Caiti[2], N Battistini[2] and G Bedogni[2,3]*

[1]*Malattie Metaboliche III e Laboratorio di Endocrinologia Sperimentale, Istituto Auxologico Italiano IRCCS, Piancavallo, Italy;* [2]*Cattedra di Nutrizione Umana, Università di Modena e Reggio Emilia, Italy; and* [3]*Centro Studi Fegato, AREA Science Park, Basovizza, Trieste, Italy*

Objective: To measure body water distribution and to evaluate the accuracy of eight-polar bioelectrical impedance analysis (BIA) for the assessment of total body water (TBW) and extracellular water (ECW) in severe obesity.
Design: Cross-sectional study.
Setting: Obesity clinic.
Subjects: In all, 75 women aged 18–66 y, 25 with body mass index (BMI) between 19.1 and 29.9 kg/m^2 (ie not obese), 25 with BMI between 30.0 and 39.9 kg/m^2 (ie class I and II obese), and 25 with BMI between 40.0 and 48.2 kg/m^2 (ie class III obese).
Methods: TBW and ECW were measured by ^2H_2O and Br dilution. Body resistance (R) was obtained by summing the resistances of arms, trunk and legs as measured by eight-polar BIA (InBody 3.0, Biospace, Seoul, Korea). The resistance index at a frequency of x kHz (RI_x) was calculated as height $^2/R_x$.
Results: ECW : TBW was similar in women with class III (46 ± 3%, mean ± s.d.) and class I–II obesity (45 ± 3%) but higher than in nonobese women (39 ± 3%, $P<0.05$). In a random subsample of 37 subjects, RI_{500} explained 82% of TBW variance ($P<0.0001$) and cross-validation of the obtained algorithm in the remaining 38 subjects gave a percent root mean square error (RMSE%) of 5% and a pure error (PE) of 2.1 l. In the same subjects, RI_5 explained 87% of ECW variance ($P<0.0001$) and cross-validation of the obtained algorithm gave a RMSE% of 8% and a PE of 1.4 l. The contribution of weight and BMI to the prediction of TBW and ECW was nil or negligible on practical grounds.
Conclusions: ECW : TBW is similar in women with class I–II and class III obesity up to BMI values of 48.2 kg/m^2. Eight-polar BIA offers accurate estimates of TBW and ECW in women with a wide range of BMI (19.1–48.2 kg/m^2) without the need of population-specific formulae.
Sponsorship: Progetti di Ricerca Corrente, Istituto Auxologico Italiano.
European Journal of Clinical Nutrition (2005) **59**, 155–160. doi:10.1038/sj.ejcn.1602049
Published online 1 September 2004

Keywords: obesity; total body water; extracellular water; bioelectrical impedance analysis

Introduction

As compared to normal-weight women, obese women have a lower total body water (TBW) per unit of weight (Wt) and a higher extracellular water (ECW) per unit of TBW (Waki *et al*, 1991). This ECW : TBW expansion persists after weight loss and may be an intrinsic feature of obesity (Mazariegos *et al*, 1992; Van Marken Lichtenbelt & Fogelholm, 1999). Most of the data on body water distribution of obese subjects were obtained in women with class I or II obesity and less data are available for women with class III obesity. Because ECW : TBW is higher in adipocytes than in other cells (Wang & Pierson, 1976), we hypothesized that ECW : TBW may be

*Correspondence: G Bedogni, Department of Hygiene, Microbiology and Biostatistics, Faculty of Medicine and Surgery, University of Modena and Reggio Emilia, Via Campi 287, 41100 Modena, Italy.
E-mail: giorgiobedogni@libero.it
Guarantors: A Sartorio and G Bedogni.
Contributors: AS coordinated the study and contributed to the final version of the manuscript; GB analyzed the data and wrote the first draft of the manuscript; MM, OC and NB performed laboratory measurements; FA and PG Marinone performed clinical measurements.
Received 14 January 2004; revised 24 June 2004; accepted 14 July 2004; published online 1 September 2004

higher in class III than in class I or II obesity. The expanded ECW : TBW of obese women may have prognostic and clinical implications (Bedogni *et al*, 2003a). For instance, the expanded ECW : TBW of obese subjects has been supposed to play a role in the pathogenesis of hypertension (Raison *et al*, 1986). The altered body water distribution of obese women has nonetheless implications for the assessment of body composition from bioelectrical impedance analysis (BIA) (Deurenberg, 1996; Steijaert *et al*, 1997). The distribution between ECW and intracellular water is in fact a primary determinant of body resistance (Deurenberg *et al*, 1989). Eight-polar BIA is a recently introduced technique with three interesting characteristics: (1) the use of very practical tactile electrodes for measuring segmental resistances at multiple frequencies, (2) the absence of need to standardize subject's posture before analysis and, (3) the rapidity of measurements. We have shown that eight-polar BIA offers accurate estimates of TBW and appendicular body composition in adult and elderly subjects (Bedogni *et al*, 2002; Malavolti *et al*, 2003). However, the accuracy of eight-polar BIA in detecting TBW and ECW in conditions of altered body water distribution is at present unknown.

The aim of this study was twofold: (1) to establish whether ECW : TBW differs in class III obesity *vs* class I–II obesity, and (2) to evaluate the accuracy of eight-polar BIA in the assessment of TBW and ECW in obese women.

Materials and methods

Subjects

The study subjects were: (1) 25 women with body mass index (BMI) between 19.1 and 29.9 kg/m^2 (ie not obese), (2) 25 women with BMI between 30.0 and 39.9 kg/m^2 (ie class I and II obese) and, (3) 25 women with BMI between 40.0 and 48.2 kg/m^2 (ie class III obese). Women with BMI $<30.0\,kg/m^2$ were recruited among the personnel of Modena University and those with BMI $\geqslant 30.0\,kg/m^2$ among the in-patients of the Third Division of Metabolic Diseases of the Italian Auxological Institute. The subjects were selected on the basis of the following criteria: (1) age $\geqslant$18 y; (2) absence of heart, kidney, liver, endocrine and other major disease; (3) menstrual cycle between the 6th and 10th day for fertile women; (4) absence of clinically detectable fluid retention (peripheral edema or lymphedema), (5) no use of drugs known to interfere with body water homeostasis and, (6) absence of vigorous physical exercise in the previous 48 h. The study protocol was approved by the Ethical Committee of the Italian Auxological Institute and all subjects gave their informed consent.

Anthropometry

Wt and height (Ht) were measured to the nearest 0.1 kg and 0.001 m following the *Anthropometric Standardization Reference Manual* (Lohman *et al*, 1988). BMI was calculated as Wt (kg)/Ht $(m)^2$.

Eight-polar BIA

Resistance (*R*) of arms, trunk and legs was measured after an overnight fast ($\geqslant$8 h) at frequencies of 5, 50, 250 and 500 kHz with an eight-polar tactile-electrode impedance-meter (InBody 3.0, Biospace, Seoul, Korea). This instrument makes use of eight tactile electrodes: two are in contact with the palm (E1, E3) and thumb (E2, E4) of each hand and two with the anterior (E5, E7) and posterior aspects (E6, E8) of the sole of each foot (Figure 1).

The subject stands with her or his soles in contact with the foot electrodes and grabs the hand electrodes. The sequence of measurements, controlled by a microprocessor, proceeds as follows. An alternating current (a.c.) of 250 μA of intensity (*I*) is applied between E1 and E5. The recorded voltage difference (*V*) between E2 and E4 is divided for I to obtain the resistance of right arm (R_{RA}). The same operation is performed with *V* recorded between E4 and E8 to obtain trunk resistance (R_T) and with *V* recorded between E6 and E8

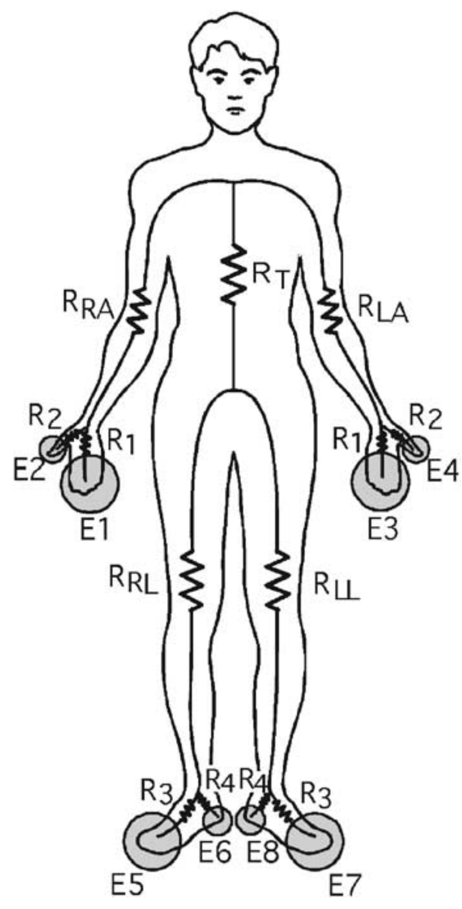

Figure 1 Measurement pathways of InBody 3.0 (reproduced with permission from Biospace). The subject stands with her or his soles in contact with the foot electrodes and grabs the hand electrodes. Abbreviations: R_{RA} = resistance of right arm; R_T = resistance of trunk; R_{LA} = resistance of left arm; R_{RL} = resistance of right leg; R_{LL} = resistance of left leg (see text for details).

to obtain the resistance of right leg (R_{RL}). The a.c. is then applied between E3 and E7 and the value of V measured between E2 and E4 is used to calculate the resistance of left arm (R_{LA}). Lastly, the value of V measured between E6 and E8 is used to calculate the resistance of left leg (R_{LL}).

Body resistance at frequency x (R_x) was calculated as the sum of segmental resistances ($R_{RA}+R_{LA}+R_T+R_{RL}+R_{LL}$). The resistance index at frequency x (RI_x) was calculated as Ht $(cm)^2/R_x$ (Ω). We avoided measuring segmental lengths and calculating segmental RIs because the above approach is at least as accurate as the segmental one and less time consuming (Bedogni *et al*, 2002; Malavolti *et al*, 2003). No caution was taken to standardize the subject's posture before BIA, as suggested by the manufacturer. Because the distance between the foot electrodes of InBody 3.0 is fixed, the legs of severely obese subjects may come in contact with each other. To establish the practical significance of this apparent violation of conduction pathways, we tested whether the TBW-RI_x and ECW-RI_x relationships did differ in obese subjects with leg-to-leg contact *vs* those without leg-to-leg contact. The regression lines had similar slopes and intercepts (data not shown), confirming the manufacturer's indication that this apparent violation of the conduction pathways is expected to have a minimal effect on the assessment of body composition. The within-day precision of InBody 3.0 in three obese subjects was $\leqslant 2.0\,\Omega$, a value similar to that observed in nonobese subjects (Bedogni *et al*, 2002; Malavolti *et al*, 2003).

^2H_2O and Br dilution

TBW was measured by ^2H_2O dilution and ECW by Br dilution. Each fasting subject ($\geqslant$8 h) received an accurately weighed solution made up of ^2H_2O, NaBr, and drinkable water. In two unselected obese subjects, ^2H_2O and Br reached the equilibrium in plasma between 3.5 and 4.0 h after administration. Plasma samples were thus collected before the administration of the solution and 4 h later. Subjects refrained from eating and drinking during the equilibration period. ^2H_2O concentration in plasma was measured by FT-IR spectrophotometry using the method of Lukaski and Johnson (1985). TBW was calculated as (^2H_2O dilution space $\times 0.95$), taking into account nonaqueous distribution of ^2H_2O. Br concentration was measured by HPLC using the method of Wong *et al* (1989). ECW was calculated as (Br dilution space $\times 0.90 \times 0.95$), taking into account nonextracellular distribution of Br and Donnan's effect, respectively. All samples were measured in triplicate and the coefficient of variation of ^2H_2O and Br measurements was $\leqslant$2%.

Statistical analysis

Sample size was determined by considering that a sample of 25 subjects has a power of 0.99 to detect a slope of 1.00 at an alpha level of 0.05 when the standard deviation of TBW is 4 l and that of RI_{500} is 3 Ω (Bedogni *et al*, 2002). Aiming at comparing nonobese, class I–II obese, and class III obese women, we enrolled 75 (25*3) subjects. Between-group comparisons were performed by ANOVA using the Fisher' s PLSD test for *post hoc* analysis. Because ANCOVA did not detect any influence of obesity status (nonobese *vs* obese class I–II *vs* obese class III, $P \geqslant 0.655$ and nonobese *vs* obese class I–II–III, $P \geqslant 0.353$) on the TBW-RI_x or ECW–TBW-RI_x relationships, we randomly split the study sample into two halves. One half ($n=37$) was used to develop predictive algorithms of TBW and ECW that were then cross-tested on the remaining half ($n=38$). The adjusted determination coefficient (R^2_{adj}), the root mean square error (RMSE) and the percent root mean square error (RMSE% = RMSE/mean value of Y) obtained from linear regression of TBW or ECW *vs* RI_x were used to determine the accuracy of BIA. In the cross-validation sample, the pure error (PE) of the estimate was also calculated. Measured and predicted values of TBW and ECW were compared using paired *t*-tests. Statistical significance was set to a value of $P<0.05$ for all tests. Statistical analysis was performed on a MacOS computer using the Statview 5.1 and SuperANOVA 1.11 software packages (SAS, Cary, NC, USA).

Results

The measurements of the women are given in Table 1.

Age was similar in nonobese, class I–II obese, and class III obese women. As expected, Wt and BMI were significantly higher in obese than in nonobese women. TBW was significantly higher in obese than in nonobese women but there was no difference in TBW between class I–II and class III obesity. However, TBW : Wt was significantly lower in women with class III obesity than in those with class I–II obesity and in these latter than in nonobese women. ECW was significantly higher in obese than in nonobese women but there was no difference in ECW between class I-II and

Table 1 Measurements of the women

	Non obese	*Obese class I–II*	*Obese class III*
n	*25*	*25*	*25*
Age (y)	39 ± 12^a	43 ± 14^a	44 ± 13^a
Wt (kg)	62.2 ± 9.7^a	93.7 ± 9.3^b	105.6 ± 12.3^c
Height (m)	1.64 ± 0.06^a	1.59 ± 0.06^b	1.56 ± 0.07^b
BMI (kg/m^2)	23.0 ± 3.4^a	36.9 ± 2.3^b	43.1 ± 2.2^c
TBW (l)	38.3 ± 3.4^a	40.9 ± 4.4^b	41.6 ± 4.4^b
TBW : wt (%)	62 ± 7^a	44 ± 4^b	40 ± 3^c
ECW (l)	15.1 ± 2.6^a	18.6 ± 3.3^b	19.1 ± 3.2^b
ECW : TBW (%)	39 ± 3^a	45 ± 3^b	46 ± 3^b
R_5 (Ω)	1487 ± 155^a	1280 ± 118^b	1207 ± 122^b
R_{500} (Ω)	1140 ± 118^a	959 ± 94^b	900 ± 90^b

Values are given as mean $\pm$ s.d. Abbreviations: BMI = body mass index; TBW = total body water; ECW = extracellular water; R_x = body resistance at x kHz; Wt = weight.

[a,b,c]Values not sharing the same superscript are significantly different at the $P<0.05$ level (Fisher's PLSD).

class III obesity. Likewise, ECW:TBW was significantly higher in obese than in nonobese women but there was no difference in ECW:TBW between class I–II and class III obesity. R_5 and R_{500} showed the same trend of ECW and TBW, being higher in non-obese than in obese women and similar in class I–II and class III obesity.

Because ANCOVA did not detect any influence of obesity status on the TBW–RI_x or ECW-RI_x relationships (see under Statistical analysis), we randomized the study sample in two halves. One-half ($n=37$) was used to develop predictive algorithms of TBW and ECW that were cross-tested on the remaining half ($n=38$).

As in our previous study (Bedogni *et al*, 2002), we developed an equation for predicting TBW from RI_{500} (panel a1 of Figure 2). RI_{500} explained 82% of TBW variance ($P<0.0001$) and the RMSE% was 5%. The mean±s.d. bias of this equation was 0.0±2.0 l, corresponding to a measured value of TBW of 40.6±4.8 l *vs* an estimated one of 40.6±4.4 l ($P=0.361$). The residuals of the TBW–RI_{500} regression were uncorrelated with Wt ($P=0.968$) and BMI ($P=0.838$). For a more direct comparison, it may be noted that RI_{500} explained 36% more variance of TBW than Wt ($R^2_{adj}=0.46$, $P<0.0001$). The cross-validation of the TBW equation yielded a RMSE% of 5% and a PE 2.1 l (panel a2 of Figure 2). The mean±s.d. bias associated with the cross-validation was 0.0±2.1 l, corresponding to a measured value of TBW of 39.9±3.7 l *vs* an estimated one of 39.9±3.4 l ($P=0.284$).

As expected by electrical theory, the most accurate prediction of ECW from RI was obtained at 5 kHz, that is, the lowest frequency measured by eight-polar BIA. RI explained in fact from 2 to 3% more variance of ECW at 5 kHz than at higher frequencies (data not shown). The predictive equation of ECW from RI_5 is given in panel b1 of Figure 2. RI_5 explained 87% of ECW variance ($P<0.0001$) and the RMSE% was 8%. The mean±s.d. bias of this equation was 0.0±1.4 l, corresponding to a measured value of ECW of 18.0±3.9 l *vs* an estimated one of 18.0±3.7 l

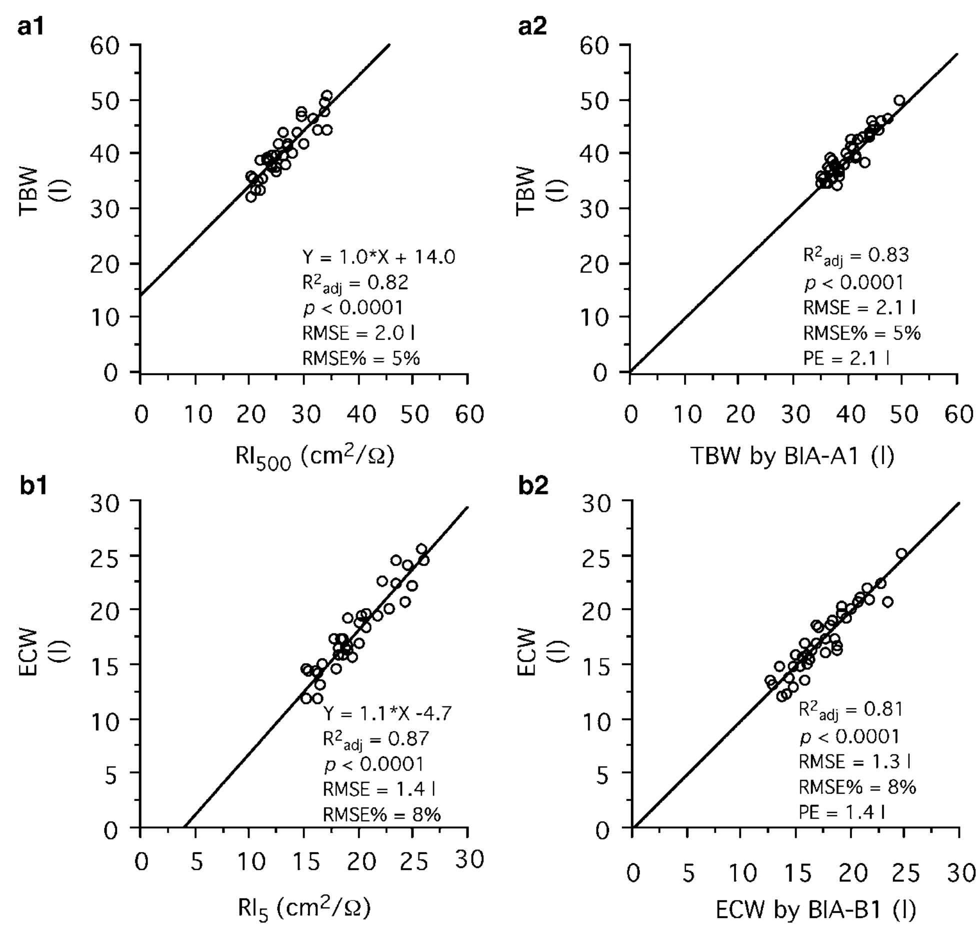

Figure 2 Generation (a1 and b1, $n=37$) and cross-validation (a2 and b2, $n=38$) of predictive equations for total body water and extracellular water in two random subsamples of women. Abbreviations: TBW = total body water; ECW = extracellular water; RI_x = resistance index at a frequency of x kHz; BIA = bioelectrical impedance analysis; R^2_{adj} = adjusted determination coefficient; RMSE = root mean squared error; RMSE% = percent root mean squared error; PE = pure error.

($P = 0.968$). The residuals of the ECW-RI_5 regression were uncorrelated with Wt ($P = 0.061$) but were associated with BMI ($R^2_{adj} = 0.19$, $P = 0.008$). For a more direct comparison, it may be noted that RI_5 explained 26% more variance of ECW than Wt ($R^2_{adj} = 0.61$, $P < 0.0001$) and 49% more than BMI ($R^2_{adj} = 0.38$, $P < 0.0001$). Inclusion of BMI as a predictor together with RI_5 did not however improve the accuracy of the estimate, as judged by an unchanged RMSE% (8%). The cross-validation of the ECW equation yielded an RMSE% of 8% and a PE of 1.4 l (panel b2 of Figure 2). The mean ± s.d. bias associated with the cross-validation was 0.0 ± 1.3 l, corresponding to a measured value of ECW of 17.2 ± 3.0 l *vs* an estimated one of 17.2 ± 2.7 l ($P = 0.564$).

Discussion

In this study, we found no difference in ECW : TBW between class III and class I–II obesity even if obese women had an expanded ECW : TBW as compared to nonobese women. Because the highest BMI of our women was one of 48.2 kg/m^2, we cannot exclude that an expansion of ECW : TBW may occur at higher levels of class III obesity as compared to class I and II obesity (Mazariegos *et al*, 1992; Guida *et al*, 2003). However, as far as values of BMI up to 48.2 kg/m^2 are considered, ECW : TBW appears to be the same in class I–II and class III obese women.

As the prediction of TBW and ECW from eight-polar BIA is concerned, this study shows that common predictive algorithms can be employed in obese and nonobese women, at least as values of BMI up to 48.2 kg/m^2 are considered. BMI had in fact no influence on the TBW–RI_{500} relationship and its contribution to the ECW–RI_{500} relationship was modest (19%). While this latter finding suggests that obesity may affect the estimate of ECW from eight-polar BIA, BMI was a less powerful predictor than RI_5 and did not add to the prediction of ECW from BIA. BMI is however just a surrogate index of body composition, especially in nonobese individuals, and accurate measurements of body fat are needed to test the influence of body composition on the TBW–BIA and ECW–BIA relationships.

The TBW equation obtained in this study has virtually the same slope (1.0 *vs* 1.1) of that obtained in a previous study of 50 nonobese subjects of both sexes (Bedogni *et al*, 2002). However, the intercepts are different (14.0 *vs* 11.1), possibly because only women with a greater interindividual variability in BMI were considered in the present study. The nil or negligible effects of Wt and BMI on the estimates of water compartments suggest nonetheless that eight-polar BIA may be less population-specific than four-polar BIA. However, a direct comparison of the two methods is needed to test this hypothesis.

We consider of great importance the fact that BMI and Wt contributed nothing or little to the variance of TBW and ECW unexplained by RI. A critique often raised to BIA is in fact that anthropometric indicators may be better predictors of body composition. We have the policy of systematically testing this hypothesis in our BIA studies. For instance, we have not been able to show that four-polar BIA at 50 kHz is more accurate than Wt in estimating TBW and appendicular body composition in anorexic women (Scalfi *et al*, 1997; Bedogni *et al*, 2003b). Even if the use of multifrequency instruments may change this evidence, the use of an impedance-meter is clearly not justified in these circumstances. However, the present study and our experience so far (Malavolti *et al*, 2003) suggest that eight-polar BIA is substantially better than Wt as an indicator of body composition. As we have suggested elsewhere (Malavolti *et al*, 2003), unique characteristics of eight-polar BIA that may contribute to its very low dependency from Wt are: (1) the use of tactile electrodes, avoiding problems connected with adhesive electrodes, (2) the fact that whole-body eight-polar R is the sum of segmental resistances obtained with a 5-cylinder model of the human body (Figure 1) and, (3) the insensitivity of eight-polar BIA to subject's posture.

Interestingly, the RMSE% and the PE associated with the cross-validation of the TBW equation in the present study were lower than in our previous study (5 *vs* 8% and 2.1 *vs* 3.1 l). Besides the larger number of subjects, it is likely that the greater interindividual variability in TBW : Wt observed in the present study has contributed to this result (Scalfi *et al*, 1997). Not unexpectedly (Bedogni *et al*, 1997), the RMSE% associated with the cross-validation of the ECW equation was higher than that associated with the TBW equation (8 *vs* 5%). However, such a RMSE% is still acceptable for field studies of body composition. The fact that RI explained more variance of ECW at 5 kHz than at higher frequencies is in agreement with electrical theory. However, the increase in the explained variance of ECW was only ⩽3% so that the inclusion of frequencies <5 kHz in the spectrum of eight-polar BIA may increase its ability to estimate ECW.

In conclusion, ECW : TBW did not differ in women with class III and class I–II obesity and eight-polar BIA offered accurate estimates of TBW and ECW in women with a wide range of BMI (19.1–48.2 kg/m^2) without the need of population-specific algorithms.

References

Bedogni G, Bollea MR, Severi S, Trunfio O, Manzieri AM & Battistini N (1997): The prediction of total body water and extracellular water from bioelectric impedance in obese children. *Eur. J. Clin. Nutr.* **51**, 129–133.

Bedogni G, Malavolti M, Severi S, Poli M, Mussi C, Fantuzzi AL & Battistini N (2002): Accuracy of an eight-point tactile-electrode impedance method in the assessment of total body water. *Eur. J. Clin. Nutr.* **56**, 1143–1148.

Bedogni G, Borghi A & Battistini N (2003a): Body water distribution and disease. *Acta Diabetol.* **40** (Suppl 1), S200–S202.

Bedogni G, Marra M, Bianchi L, Malavolti M, Nicolai E, De Filippo E & Scalfi L (2003b): Comparison of bioelectrical impedance analysis and dual-energy X-ray absorptiometry for the assessment of appendicular body composition in anorexic women. *Eur. J. Clin. Nutr.* **57**, 1068–1072.

Deurenberg P (1996): Limitations of the bioelectrical impedance method for the assessment of body fat in severe obesity. *Am. J. Clin. Nutr.* **64**, 449S–452S.

Deurenberg P, van der Kooy K, Leenen R & Schouten FJ (1989): Body impedance is largely dependent on the intra- and extra-cellular water distribution. *Eur. J. Clin. Nutr.* **43**, 845–853.

Guida B, Trio R, Pecoraro P, Gerardi MC, Laccetti R, Nastasi A & Falconi C (2003): Impedance vector distribution by body mass index and conventional bioelectrical impedance analysis in obese women. *Nutr. Metab. Cardiovasc. Dis.* **13**, 72–79.

Lohman T, Roche A & Martorell R eds. (1988): *Anthropometric Standardization Reference Manual*. Champaign, IL: Human Kinetics Books.

Lukaski HC & Johnson PE (1985): A simple, inexpensive method of determining total body water using a tracer dose of D_2O and infrared absorption of biological fluids. *Am. J. Clin. Nutr.* **41**, 363–370.

Malavolti M, Mussi C, Poli M, Fantuzzi AL, Salvioli G, Battistini N & Bedogni G (2003): Cross-calibration of eight-polar bioelectrical impedance analysis versus dual-energy X-ray absorptiometry for the assessment of total and appendicular body composition in healthy subjects aged 21–82 years. *Ann. Hum. Biol.* **30**, 380–391.

Mazariegos M, Kral JG, Wang J, Waki M, Heymsfield SB, Pierson RN, Thornton JC & Yasumura S (1992): Body composition and surgical treatment of obesity. Effects of weight loss on fluid distribution. *Ann. Surg.* **216**, 69–73.

Raison J, Achimastos A, Asmar R, Simon A & Safar M (1986): Extracellular and interstitial fluid volume in obesity with and without associated systemic hypertension. *Am. J. Cardiol.* **57**, 223–226.

Scalfi L, Bedogni G, Marra M, Di Biase G, Caldara A, Severi S, Contaldo F & Battistini N (1997): The prediction of total body water from bioelectrical impedance in patients with anorexia nervosa. *Br. J. Nutr.* **78**, 357–365.

Steijaert M, Deurenberg P, Van Gaal L & De Leeuw I (1997): The use of multi-frequency impedance to determine total body water and extracellular water in obese and lean female individuals. *Int. J. Obes. Relat. Metab. Disord.* **21**, 930–934.

Van Marken Lichtenbelt WD & Fogelholm M (1999): Increased extracellular water compartment, relative to intracellular water compartment, after weight reduction. *J. Appl. Physiol.* **87**, 294–298.

Waki M, Kral JG, Mazariegos M, Wang J, Pierson RN & Heymsfield SB (1991): Relative expansion of extracellular fluid in obese vs. *nonobese women. Am. J. Physiol.* **261**, E199–E203.

Wang J & Pierson RN (1976): Disparate hydration of adipose and lean tissue requires a new model for body water distribution in man. *J. Nutr.* **106**, 1687–1693.

Wong WW, Sheng HP, Morkeberg JC, Kosanovich JL, Clarke LL & Klein PD (1989): Measurement of extracellular water volume by bromide ion chromatography. *Am. J. Clin. Nutr.* **50**, 1290–1294.