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ORIGINAL ARTICLE

Cross-validation of bioelectrical impedance analysis for the assessment of body composition in a representative sample of 6- to 13-year-old children

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Background/Objectives: (1) To cross-validate tetra- (4-BIA) and octopolar (8-BIA) bioelectrical impedance analysis vs dual-energy X-ray absorptiometry (DXA) for the assessment of total and appendicular body composition and (2) to evaluate the accuracy of external 4-BIA algorithms for the prediction of total body composition, in a representative sample of Swiss children.
Subjects/Methods: A representative sample of 333 Swiss children aged 6–13 years from the Kinder-Sportstudie (KISS) (ISRCTN15360785). Whole-body fat-free mass (FFM) and appendicular lean tissue mass were measured with DXA. Body resistance (R) was measured at 50 kHz with 4-BIA and segmental body resistance at 5, 50, 250 and 500 kHz with 8-BIA. The resistance index (RI) was calculated as height^2/R. Selection of predictors (gender, age, weight, RI4 and RI8) for BIA algorithms was performed using bootstrapped stepwise linear regression on 1000 samples. We calculated 95% confidence intervals (CI) of regression coefficients and measures of model fit using bootstrap analysis. Limits of agreement were used as measures of interchangeability of BIA with DXA.
Results: 8-BIA was more accurate than 4-BIA for the assessment of FFM (root mean square error (RMSE) = 0.90 (95% CI 0.82–0.98) vs 1.12 kg (1.01–1.24); limits of agreement 1.80 to -1.80 kg vs 2.24 to -2.24 kg). 8-BIA also gave accurate estimates of appendicular body composition, with RMSE $\leqslant$0.10 kg for arms and $\leqslant$0.24 kg for legs. All external 4-BIA algorithms performed poorly with substantial negative proportional bias ($r \geqslant 0.48$, $P < 0.001$).
Conclusions: In a representative sample of young Swiss children (1) 8-BIA was superior to 4-BIA for the prediction of FFM, (2) external 4-BIA algorithms gave biased predictions of FFM and (3) 8-BIA was an accurate predictor of segmental body composition.
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Keywords: epidemiology; body composition; bioelectrical impedance analysis; dual-energy X-ray absorptiometry; prediction equations

Introduction

The amount and distribution of body fat and the amount and composition of fat-free mass (FFM) are associated with important health outcomes in infancy, childhood and later adulthood (Wells, 2003; Wells and Fewtrell, 2006). The measurement of body composition is thus considered a central outcome in current clinical and epidemiological pediatric research (Pietrobelli, 2004).

However, reference methods for the assessment of body composition, such as the four-compartment model, are not suitable for use in epidemiological studies because of their complexity and cost (Wells and Fewtrell, 2006). Although not as accurate as the four-compartment model, dual-energy X-ray absorptiometry (DXA) compares well with reference methods, and is increasingly used to calibrate indirect techniques (Malavolti *et al.*, 2003; Kim *et al.*, 2006; Lohman and Going, 2006). An attractive feature of DXA is the

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possibility of obtaining measurements of appendicular body composition (Gallagher *et al*., 1997; Malavolti *et al*., 2003). These measurements are useful to track changes in segmental bone and soft tissues occurring with normal and pathological growth (Fuller *et al*., 2002; Chomtho *et al*., 2006). However DXA is expensive, lacks portability and exposes to ionizing radiations. Thus, researchers look for indirect methods which are cheaper, more portable and less invasive for children (Guo *et al*., 1996; Wells and Fewtrell, 2006).

Among indirect methods, bioelectrical impedance (BIA) is presently the best option for the assessment of FFM and appendicular muscle mass (Malavolti *et al*., 2003; Pietrobelli *et al*., 2004). BIA is usually performed using a single- or multifrequency tetrapolar (4-BIA) technique with adhesive electrodes (Kushner, 1992). An octopolar (8-BIA) multifrequency implementation of BIA with tactile electrodes was recently made available on the market (Bedogni *et al*., 2002). Contrarily to 4-BIA (Bedogni *et al*., 2003b; Bertoli *et al*., 2007), 8-BIA was consistently found to contribute more than anthropometry to the prediction of body composition (Bedogni *et al*., 2002; Malavolti *et al*., 2003; Medici *et al*., 2005). However, 8-BIA has not been directly compared to 4-BIA and has been validated only for selected physiological and clinical conditions (Bedogni *et al*., 2002; Malavolti *et al*., 2003; Medici *et al*., 2005; Sartorio *et al*., 2005). Moreover, all 4-BIA algorithms currently available for children were developed in convenience samples (Nielsen *et al*., 2007).

The Kinder-Sportstudie (KISS) is a randomized controlled trial aimed to test whether an extended physical activity program improves physical activity, physical fitness, body composition and quality of life in a representative sample of Swiss children aged 6–13 years (Zahner *et al*., 2006). The KISS study offered us the unique opportunity to cross-calibrate 4-BIA and 8-BIA against DXA in a representative sample of the general pediatric population. The aims of the present study were (1) to cross-calibrate 4-BIA and 8-BIA against DXA for the assessment of total and appendicular body composition and (2) to evaluate the accuracy of external BIA algorithms for the prediction of total body composition, in a representative sample of Swiss children.

Materials and methods

Subjects and study protocol

The study design of KISS (ISRCTN15360785) was reported in detail elsewhere (Zahner *et al*., 2006). The KISS children were randomly selected and stratified by class, geographic area and ethnicity to be representative of Swiss children with respect to gender, sociodemographic status and body mass index (BMI). Being a randomized controlled trial, KISS included a baseline and a follow-up visit after 1 academic year. The KISS baseline data were used for the present analysis. Informed consent for all measurements was given by each child and a parent. The study was approved by the ethics committees of the University of Basel, the ETH of Zürich, as well as by the cantonal ethical committee of Aargau, Switzerland.

A total of 497 out of 502 children had complete general data and 366 of them (74%) had undergone anthropometry, DXA, 4-BIA and 8-BIA measurements and were evaluated for the present analysis. All measurements besides DXA were performed at school. The lack of DXA data for 131 children (26%) was due to the request of their parents that they were not exposed to ionizing radiations. The option to refuse DXA measurements was in fact systematically offered during KISS. DXA, anthropometry and BIA were performed within 2 days. 4-BIA and 8-BIA measurements were taken within 60 min. The children arrived at school after an overnight fast. After blood drawing and before BIA, the children ate a small breakfast made of one roll and about 200 ml of apple or orange juice. It was in fact considered ethically unjustifiable to let the fasting proceed another 2 h before BIA measurements. This small quantity of food and liquids is however not expected to affect body impedance (Kushner *et al*., 1996), as also shown by studies performed in our laboratory on adult subjects (RF Kushner, R Gudivaka and DA Schoeller, unpublished data).

Anthropometry and pubertal status

Body weight and height were measured by the same operator following the *Anthropometric Standardization Reference Manual* (Lohman *et al*., 1988). BMI was calculated as weight (kg)/height $(m)^2$. To allow international comparisons and thus for descriptive purposes only, we calculated *z*-scores of BMI using US reference data (Kuczmarski *et al*., 2000). Pubertal status was self-assessed using Tanner' s criteria (Tanner, 1962).

4-BIA

Whole-body resistance (R4) was measured by the same two operators with a tetrapolar adhesive-electrode impedance meter (model 101A; RJL Systems, Detroit, MI, USA) at a frequency of 50 kHz following NIH guidelines (NIH, 1996). When the distance between the proximal and distal electrode was below 5 cm, the proximal electrode was moved more cranial to achieve the recommended distance of 5 cm. The coefficient of variation (CV) for repeated within-day 4-BIA measurements with replacement of electrodes was 2.0%. The whole-body tetrapolar resistance index (RI4) was calculated as height $(cm)^2$/R4 (Ω) (Kushner, 1992).

8-BIA

Resistance (R8) of arms, trunk and legs was measured by the same operator with an octopolar tactile-electrode impedance meter (InBody 3.0, Biospace, Seoul, Korea) at frequencies of 5, 50, 250 and 500 kHz following the manufacturer's guidelines. 8-BIA makes use of eight tactile electrodes: two are in

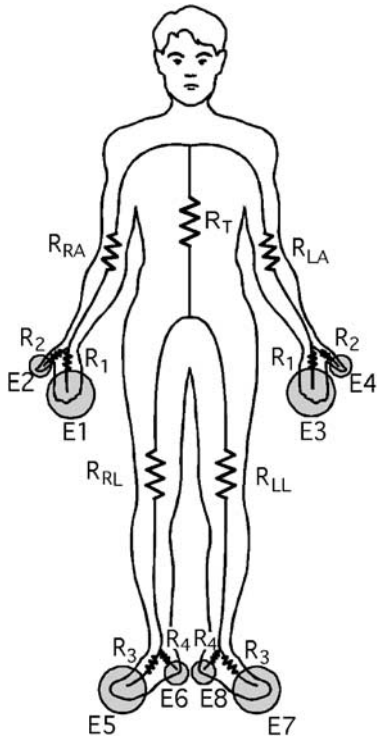

Figure 1 Current pathways for octopolar bioelectrical impedance (BIA; graph reproduced with permission of Biospace). R_{RA} = resistance of right arm; R_T = resistance of trunk; R_{LA} = resistance of left arm; R_{RL} = resistance of right leg; R_{LL} = resistance of left leg (see text for description).

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 his or her 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 of 250 μA of intensity (*I*) is applied between E1 and whole-body resistance at frequency *x* ($R8_x$, whole body) was calculated as the sum of segmental $R8_x$ (right arm + left arm + trunk + right leg + left leg). The CV for repeated within-day 8-BIA measurements at multiple frequencies was $\leqslant$2.0%. RI were calculated as height $(cm)^2/R8_x$ (Ω), where $R8_x$ is the resistance of whole-body, arm or leg at frequency *x* (Bedogni *et al*., 2002).

DXA

The three-compartment DXA model separates body mass into lean tissue mass (LTM), fat mass (FM) and bone mineral content (BMC) (Pietrobelli *et al*., 1996). The sum of LTM and BMC represents FFM and LTM is synonym with muscle mass at the appendicular level (Wang *et al*., 1996; Malavolti *et al*., 2003; Pietrobelli *et al*., 2004). DXA was performed by the same operator using a Hologic QDR-4500 densitometer coupled with pediatric software (Hologic, Waltham, MA, USA). The densitometer was calibrated daily against the standard phantom provided by the manufacturer.

Choice of external BIA algorithms for cross-validation

The criteria used to select external BIA algorithms for cross-validation in KISS children were (1) use of DXA as the reference method; (2) use of foot-to-leg BIA as the indirect method; (3) use of FFM as the outcome measure; (4) availability of data for Caucasian subjects; (5) availability of data for both genders; (6) age comparable to that of the KISS children and (7) no use of predictors other than gender, age, weight, height and BIA-based indexes. Using these criteria we identified four algorithms for testing in the KISS population (de Lorenzo *et al*., 1998; Bedogni *et al*., 2002; Pietrobelli *et al*., 2003; Nielsen *et al*., 2007).

Statistical analysis

Continuous variables are given as means and standard deviations (s.d.). Between-group comparisons were performed with unpaired Student's *t*-test for continuous variables and with Fisher's exact test for categorical variables. Selection of the variables for FFM prediction algorithms was performed by bootstrapped backward stepwise linear regression on 1000 random samples (*P*-value to enter = 0.05; *P*-value to remove = 0.1) (Harrell, 2001). The candidate predictors were gender (male = 1; female = 0), pubertal status (Tanner stage 1 = 1; Tanner stages 2–5 = 0), age, weight, RI4 at 50 kHz, and RI8 at 5, 50, 250 and 500 kHz. Predictors identified at bootstrap analysis were entered into multiple regression models with 95% confidence intervals (95% CI) calculated by bootstrap analysis on 1000 random samples. Standardized regression coefficients were calculated to quantify the independent contribution of predictors. The accuracy of the algorithms was evaluated using the adjusted coefficient of determination (R^2_{adj}) and the root mean square error (RMSE) with 95% CI calculated on the same 1000 bootstrap samples used for calculating 95% CI of regression coefficients (Efron and Tibshirani, 1993). Bland and Altman's method was used to calculate the limits of agreement between BIA algorithms and DXA, and Pitman's test was used to evaluate proportional bias (Ludbrook, 2002). Fixed bias was defined as the difference between the value estimated by BIA and that measured by DXA. Statistical significance was set to a two-tailed $P<0.05$. Statistical analysis was performed using STATA 10.0 (StataCorp, College Station, TX, USA).

Results

Of the 497 KISS children for whom complete general data were available, 366 (74%) underwent anthropometry, DXA, 4-BIA and 8-BIA measurements and were considered for the present analysis. The children without DXA measurements

were younger than those with DXA measurements (8.4 (2.1) vs 9.5 (2.1) years, mean (s.d.); $P<0.0001$) but had the same gender ($P=0.731$) and *z*-scores of weight ($P=0.196$), height ($P=0.278$) and BMI ($P=0.245$) (data not shown).

Of these 366 children, 33 (9%) had at least one BIA measurement $\geqslant 3.5$ internal s.d. scores and were classified as outliers. After having checked that the outlying values were not due to input errors, we removed these children from the data set. As 8-BIA is concerned, virtually all outlying values were related to arms and/or trunk. This led us to hypothesize that some children may not have grabbed the hand electrodes with sufficient strength (despite constant observation by a technician). As 4-BIA is concerned, the outlying values were compatible with electrode detachment. The children with outlying values did not differ in any other measurement from those with acceptable values of 4-BIA and 8-BIA. Moreover, there were no outliers for any other measurement of interest.

The measurements of the remaining 333 children are given in Table 1. Age was similar in girls ($n=171$) and boys ($n=162$), but more girls than boys had entered puberty, that is, had a Tanner stage >1 (40 vs 20%; $P<0.0001$).

Weight, height and BMI were similar in boys and girls both as absolute values and *z*-scores. FM was higher in girls in both absolute and percent terms ($P\leqslant 0.002$), while whole-body FFM and segmental LTM were higher in boys ($P\leqslant 0.029$). Coherently with the latter finding, whole-body and appendicular values of R4 and R8 were systematically lower in boys than in girls ($P<0.0001$; only values of R4 at 50 kHz and R8 at 500 kHz are given in Table 2.).

The mean (s.d.) difference between body mass measured by DXA and body weight measured by balance was 1.3 kg (0.5) ($n=333$). Even if this difference is statistically significant ($P<0.0001$), it is low and of no practical relevance.

Backward stepwise linear regression was performed on 1000 bootstrap samples of 333 subjects to identify the strongest predictors of whole-body FFM and segmental LTM (see 'Materials and methods' for details). Table 2 gives the number of times out of 1000 that the candidate predictors were selected for inclusion in the models. Age and weight were predictors of body composition in all bootstrap samples (1000) for all models. Gender was selected in 914–1000 bootstrap samples depending on the model. RI8 at 500 kHz was the most predictive BIA measurement for all models (655–984 bootstrap samples) with the surprising exception of left leg (174 bootstrap samples). However, because there was no between-leg difference in the LTM-$RI8_{500}$ relationship and 500 kHz was the most accurate frequency as a whole, we used $RI8_{500}$ for all algorithms.

The final algorithms including the best predictors (gender, age, weight and RI8 at 500 kHz) and a separate one based on RI4 at 50 kHz were cross-validated on 1000 bootstrap samples (Table 3). The algorithm for 8-BIA was more accurate than that for 4-BIA. The RMSE were 0.90 (95% CI 0.82–0.98) and 1.12 kg (95% CI 1.01–1.24) for 8- and 4-BIA, respectively, equivalent to 3.2 and 3.7% of FFM. RI contributed more than weight to the prediction of FFM with 8-BIA (standardized regression coefficient = 0.49 vs 0.39; $P<0.0001$) but not with 4-BIA (0.42 vs 0.46; $P<0.0001$). Because our 4-BIA impedance meter was single frequency, a comparison with

Table 1 Measurements of KISS children with availability of body composition measurements after exclusion of 33 outliers

	Females (n = *171*)	*Males* (n = *162*)	P[a]
Age (years)	9.5 (2.1)	9.7 (2.0)	0.499
Tanner stage (1/2–5, *n*)	102/69	129/33	<0.0001
Weight (kg)	33.2 (9.0)	33.5 (9.4)	0.749
z-Weight (s.d.)	0.1 (0.9)	0.1 (0.9)	0.694
Height (m)	1.37 (0.12)	1.38 (0.13)	0.569
z-Height (s.d.)	0.2 (0.9)	0.2 (0.8)	0.935
BMI ($kg\,m^{-2}$)	17.3 (2.5)	17.2 (2.5)	0.843
z-BMI (s.d.)	0.1 (1.0)	0.1 (0.9)	0.835
FFM (kg)[b]	25.7 (6.3)	27.4 (6.6)	0.017
FM (kg)[b]	8.8 (3.7)	7.5 (3.9)	0.002
FM% (%)[b]	24.8 (5.4)	20.7 (5.4)	<0.0001
LTM, right arm (kg)[b]	1.3 (0.3)	1.4 (0.4)	0.001
LTM, left arm (kg)[b]	1.2 (0.3)	1.4 (0.4)	0.001
LTM, right leg (kg)[b]	4.1 (1.2)	4.4 (1.3)	0.025
LTM, left leg (kg)[b]	4.1 (1.2)	4.4 (1.3)	0.029
$R4_{50}$, whole body (Ω)	694 (74)	655 (64)	<0.0001
$R8_{500}$, whole body (Ω)	1242 (128)	1154 (109)	<0.0001
$R8_{500}$, left arm (Ω)	361 (39)	334 (35)	<0.0001
$R8_{500}$, right arm (Ω)	357 (42)	328 (35)	<0.0001
$R8_{500}$, left leg (Ω)	253 (28)	238 (25)	<0.0001
$R8_{500}$, right leg (Ω)	252 (28)	236 (24)	<0.0001

Abbreviations: BMI, body mass index; FFM, fat-free mass; FM, fat mass; FM%, fat mass standardized on body mass; LTM, lean tissue mass; $R4_{50}$, resistance with tetrapolar BIA at 50 kHz; $R8_{500}$, resistance with octopolar BIA at 500 kHz; s.d., standard deviation; *z*-BMI, *z*-score of BMI; *z*-height, *z*-score of height; *z*-weight, *z*-score of weight.
Values are mean and standard deviations (in parentheses) unless specified otherwise.
[a]Fisher's exact test for Tanner stage and Student's unpaired *t*-test for other variables.
[b]Measured by dual-energy X-ray absorptiometry.

Table 2 Selection of candidate predictors of total and appendicular body composition at bootstrapped backward stepwise linear regression

	FFM Whole-body 8-BIA	*FFM Whole-body 4-BIA*	*LTM Left arm 8-BIA*	*LTM Right arm 8-BIA*	*LTM Left leg 8-BIA*	*LTM Right leg 8-BIA*
Male	**973**	**1000**	**995**	**999**	**914**	**925**
Puberty	491	628	118	384	145	127
Age	**1000**	**1000**	**1000**	**1000**	**1000**	**1000**
Weight	**1000**	**1000**	**1000**	**1000**	**1000**	**1000**
$RI4_{50}$	135	**1000**	—	—	—	—
$RI8_5$[a]	80	—	283	465	463	451
$RI8_{50}$[a]	90	—	478	302	175	235
$RI8_{250}$[a]	77	—	671	348	786	410
$RI8_{500}$[a]	**984**	—	**966**	**911**	**174**	**655**

Abbreviations: —, not tested; FFM, fat-free mass; LTM, lean tissue mass; $RI4_{50}$, tetrapolar resistance index at 50 kHz; $RI8_x$, octopolar resistance index at *x* kHz.
Values are the number of times out of 1000 that variables were selected for inclusion in the given model. Variables selected for algorithms are in bold.
[a]Whole-body or segmental value depending on the outcome variable.

Table 3 KISS algorithms for the assessment of total and appendicular body composition

	FFM	*FFM*	*LTM*	*LTM*	*LTM*	*LTM*
	Whole-body	*Whole-body*	*Left arm*	*Right arm*	*Left leg*	*Right leg*
	8-BIA	*4-BIA*	*8-BIA*	*8-BIA*	*8-BIA*	*8-BIA*
Male	0.36* (0.14 to 0.58) [0.03]	0.77** (0.49 to 1.05) [0.06]	0.04** (0.02 to 0.07) [0.06]	0.05** (0.03 to 0.07) [0.07]	0.08* (0.03 to 0.14) [0.03]	0.09** (0.04 to 0.14) [0.04]
Age	0.50** (0.42 to 0.58) [0.16]	0.46** (0.36 to 0.56) [0.14]	0.03** (0.02 to 0.04) [0.18]	0.03** (0.02 to 0.04) [0.16]	0.09** (0.07 to 0.11) [0.15]	0.10** (0.08 to 0.13) [0.17]
Weight (kg)	0.27** (0.25 to 0.30) [0.39]	0.32** (0.29 to 0.36) [0.46]	0.01** (0.01 to 0.01) [0.25]	0.01** (0.01 to 0.01) [0.28]	0.05** (0.04 to 0.06) [0.37]	0.05** (0.04 to 0.06) [0.38]
$RI8_{500}$ ($cm^2\,\Omega^{-1}$)[a]	0.88** (0.80 to 0.95) [0.49]	—	0.02** (0.01 to 0.02) [0.59]	0.02** (0.01 to 0.02) [0.57]	0.03** (0.03 to 0.04) [0.52]	0.03** (0.03 to 0.03) [0.48]
$RI4_{50}$ ($cm^2\,\Omega^{-1}$)	—	0.41** (0.35 to 0.46) [0.42]	—	—	—	—
Intercept	−1.78** (−2.25 to −1.32)	−0.77* (−1.30 to −0.24)	−0.23** (−0.28 to −0.18)	−0.22** (−0.27 to −0.16)	−1.10** (−1.22 to −0.99)	−1.03** (−1.14 to −0.92)
R^2_{adj}, model fit	0.98** (0.98 to 0.98)	0.97** (0.96 to 0.98)	0.94** (0.92 to 0.95	0.93** (0.91 to 0.94)	0.96** (0.96 to 0.97)	0.96** (0.96 to 0.97)
RMSE (kg), model fit	0.90 (0.82 to 0.98)	1.12 (1.01 to 1.24)	0.09 (0.08 to 0.09)	0.10 (0.09 to 0.11)	0.24 (0.22 to 0.26)	0.23 (0.21 to 0.25

Abbreviations: —, not tested; FFM, fat-free mass; LTM, lean tissue mass; R^2_{adj}, adjusted coefficient of determination; $RI4_{50}$, tetrapolar resistance index at 50 kHz; $RI8_{500}$, octopolar resistance index at 500 kHz; RMSE, root mean square error of the estimate.
Values for predictors are regression coefficients with bootstrapped 95% CI (in round brackets) and standardized regression coefficients (in square brackets). The 95% CI for model-fitting metrics were calculated by bootstrap analysis (see text for details).
[a]Whole body or segmental value employed depending on the outcome.
*$P<0.01$; **$P<0.001$.

multifrequency 8-BIA is somewhat unfair. However, a direct comparison of the contribution of 4-BIA and 8-BIA at 50 kHz confirmed that 8-BIA contributes more than weight to the prediction of FFM (standardized regression coefficient = 0.50 vs 0.38; $P<0.0001$; algorithm not shown). Expectedly for a study performed in a representative sample of the general population, the 8-BIA algorithms for appendicular LTM had virtually the same regression coefficients for the left and right side of the body. The percent RMSE of LTM were 6.9% for left arm, 7.3% for right arm, 5.6% for left leg and 5.4% for right leg.

Figure 2 gives Bland–Altman plots of the bias of KISS algorithms. Limits of agreement were −1.80 to 1.80 kg for FFM by 8-BIA, −2.24 to 2.24 kg for FFM by 4-BIA, −0.18 to 0.18 kg for LTM of left arm (8-BIA), −0.20 to 0.20 kg for LTM of right arm (8-BIA), −0.48 to 0.48 for LTM of left leg (8-BIA) and −0.46 to 0.46 kg for LTM of right leg (8-BIA). There was evidence of (minimal) proportional bias only for left and right arms ($r=-0.140$, $P=0.01$ and $r=-0.129$, $P=0.02$, respectively).

Table 4 gives the fixed and proportional bias associated with the use of external and internal algorithms. All external 4-BIA algorithms performed poorly, with limits of agreement comprised between −3.2 to 2.7 and −6.7 to 3.4 kg and substantial negative proportional bias ($r\geqslant 0.48$, $P<0.001$).

Discussion

KISS is the first cross-validation study of BIA performed in a representative sample of children from the general population. It is also the first study to provide a head-to-head comparison of 4-BIA and 8-BIA for the assessment of body composition.

In our previous research performed in adults, we have consistently found that RI8 contributes more than weight to the prediction of total body water, extracellular water and FFM (Bedogni *et al.*, 2002, 2003a; Malavolti *et al.*, 2003; Medici *et al.*, 2005), contrarily to what happens for RI4 (Bedogni *et al.*, 2003b; Bertoli *et al.*, 2007). The analysis of the KISS data confirms that weight contributes more than RI4 and less than RI8 to the prediction of FFM in young children. This could be due to the fact that 8-BIA might be more capable than 4-BIA to detect differences in body water distribution between extra- and intracellular spaces. As water distribution changes during maturation (Chumlea *et al.*, 2007), the multiple-frequency 8-BIA might have been able to estimate the relative proportions of intra- and extracellular water and thus correct for the different maturational levels.

Importantly, as determined by standardized regression coefficients, 8-BIA was the best independent predictor of body composition at the whole-body level and was second only to weight at the segmental level. Even if age and gender contributed substantially less than RI and weight to the prediction of total and appendicular body composition, their contribution was very similar for every body segment,

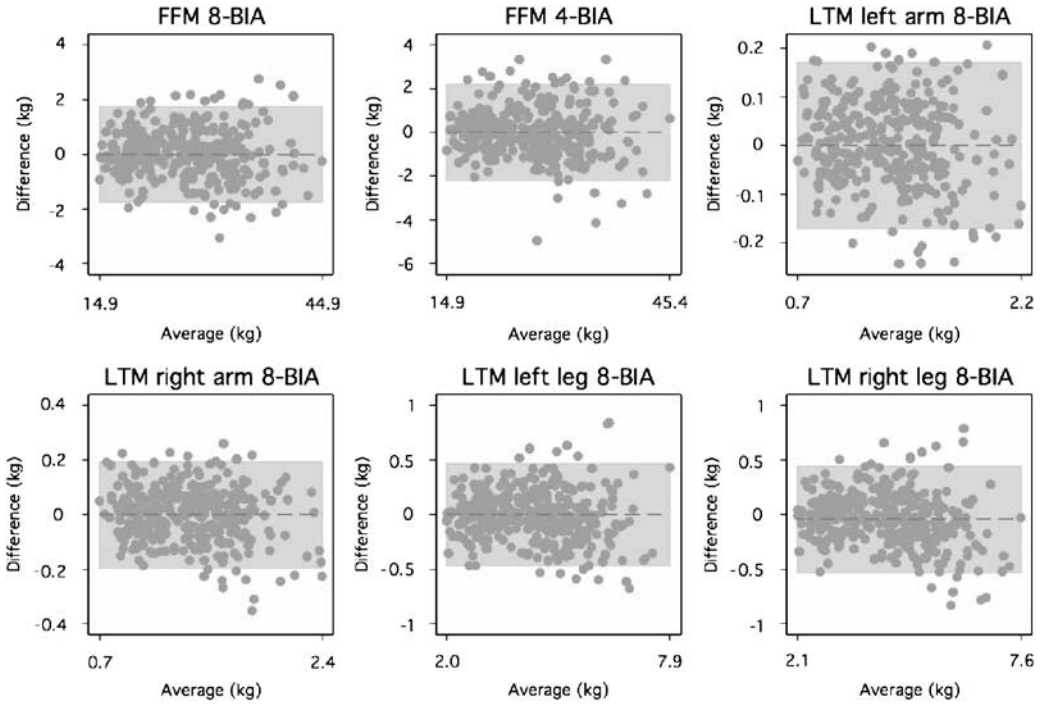

Figure 2 Bias of bioelectrical impedance analysis for the assessment of whole-body fat-free mass in Kinder-Sportstudie (KISS) children. FFM, fat-free mass; LTM, lean tissue mass; 4-BIA, tetrapolar BIA; 8-BIA, octopolar BIA. Difference (bias) was calculated as the difference between the estimate made by BIA and the measurement made by dual-energy X-ray absorptiometry (DXA).

Table 4 Bias of bioelectrical impedance analysis for the assessment of whole-body fat-free mass in KISS children

	Sample	*Fixed bias*			*Proportional bias*		
	n	*Age (years)*	*Mean (s.d.)*	P*	*Limits of agreement (kg)*	r	P**
Bedogni *et al.*, 2003a	52	8–12	1.6 (2.4)	<0.0001	−6.3 to 3.2	−0.73	<0.0001
de Lorenzo *et al.*, 1998	35	8–13	0.2 (1.5)	0.004	−3.2 to 2.7	−0.48	<0.0001
Pietrobelli *et al.*, 2003	75	7–14	1.7 (2.5)	<0.0001	−6.7 to 3.4	−0.71	<0.0001
Nielsen *et al.*, 2007	101	9–13	0.7 (2.0)	<0.0001	−4.8 to 3.3	−0.67	<0.0001
KISS 4-BIA (Table 3)	333	6–12	0.0 (1.1)	1.000	−2.2 to 2.2	−0.09	0.116
KISS 8-BIA (Table 3)	333	6–12	0.0 (0.9)	1.000	−1.8 to 1.8	−0.07	0.208

Abbreviations: BIA, bioelectrical impedance; KISS, Kinder-Sportstudie; .s.d., standard deviation.
Bias was calculated as the difference between the estimate made by BIA and the measurement made by dual-energy X-ray absorptiometry.
*Tests the null hypothesis that the mean bias equals 0.
**Tests the null hypothesis of no association between the bias and the average of the methods.

confirming that they are independent predictors of body composition in young children (Maynard *et al.*, 2001). The KISS study confirms therefore that 8-BIA can be used to assess the body composition of arms and legs of young children.

The external BIA algorithms for the assessment of FFM that we tested in KISS children had wide limits of agreement as compared to DXA and, more importantly, showed substantial negative proportional bias (thus making the use of fixed bias not useful to evaluate their accuracy). The bias may be partially due to the fact that none of these algorithms covered the entire range of age of KISS children. Another reason may be that these studies employed a Lunar densitometer as compared to the Hologic one employed by KISS (Plank, 2005). The most important message of this cross-validation of external algorithms on a representative sample of children is a reinforcement of the concept of the population specificity of BIA (Deurenberg *et al.*, 1989; Guo *et al.*, 1996; Nielsen *et al.*, 2007).

Even if KISS is the first validation study of BIA performed in a representative sample of the general population, it has

some limitations. First, 26% of KISS children did not undergo complete body composition measurements. These children were younger than those who underwent body composition measurements but had the same gender and *z*-scores of weight, height and BMI. Thus, we believe that the generalizability of our findings is not compromised by this fact. Second, 4-BIA was performed only at 50 kHz because a multifrequency tetrapolar impedance meter was not available for the KISS study. A comparison of multifrequency 8-BIA vs single-frequency 4-BIA is thus somewhat unfair. However, we proved that at the same frequency of 50 kHz, 8-BIA but not 4-BIA contributes more than anthropometry to the prediction of FFM in young children. Third, we might have also used 4-BIA to estimate appendicular body composition (Fuller and Elia, 1989; Bedogni *et al*., 2003b), but we reasoned that the time required by electrode repositioning and the higher possibility of error as compared to automatic selection of current pathways were too high to justify the use of segmental 4-BIA in an epidemiological study such as KISS.

In conclusion, in a representative sample of young Swiss children (1) 8-BIA was superior to 4-BIA for the prediction of FFM; (2) external 4-BIA algorithms gave biased predictions of FFM and (3) 8-BIA was an accurate predictor of segmental body composition.

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