TITLE PAGE

Validity of the Slaughter's Equations and Bioelectrical Impedance against Dual-energy X-ray Absorptiometry in children

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KEYWORDS

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Criterion validity of adiposity assessment methods

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DISCLOSURE

The authors declare that they do not have any conflict of interest

AUTHOR CONTRIBUTIONS

MMM, JMG, JHM, and FBO contributed to design the experiment and study design and obtained the data. MMM and FBO analysed the data. MMM and JMG drafted the manuscript under the supervision of FBO (Principal Investigator). JHM performed additional analyses, and EUG and LGM contributed in the edition and interpretation of the manuscript. All authors drafted or critically revised the manuscript for important intellectual content and approved the final version of the manuscript to be published.

BULLETS POINTS

- Few validation studies included Fat Mass Index (FMI) in children and/or adolescents
- The accuracy of Slg-Eq and BIA in measuring changes in BFP and FMI over the time is scarcely studied
- Validation of Slg-Eq and BIA against DXA in different degrees of obesity and their changes over the time is clinically relevant

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ABSTRACT

Objectives: i) To analyze the criterion validity of Slaughter's equations (Slg-Eq) and bioelectrical impedance analysis (BIA) to estimate body fat percentage (BFP) and fat mass index (FMI) at different degrees of obesity in children, compared to dual-energy X-ray absorptiometry (DXA); and ii) to determine their agreement over the time.

Methods: 92 children with overweight/obesity (10.0±1.2 years; 34.8% girls) participated in this 20-week study. Anthropometric, BIA and DXA measurements were performed.

Results: i) Both Slg-Eq and BIA methods underestimated BFP and FMI against DXA and the bias was markedly larger with BIA (Mean Absolute Percentage Error-MAPE=11% for Slg-Eq vs. 18-21% for BIA); a larger underestimation was observed in girls compared to boys for Slg-Eq (P≤0.001), and the observed underestimation in adiposity was reduced as weight status increased. ii) Systematic errors were kept constant over the time, so that no large differences between methods were observed in the change in adiposity.

Conclusions: At the group level, Slg-Eq provides a more valid estimation of BFP and FMI than BIA. At the individual level, Slg-Eq shows larger estimation errors. The validity of these methods might differ in sex and weight status. Nevertheless, both methods seem to be valid for monitoring changes in adiposity.

Keywords: Body composition, Anthropometry, Degree of obesity, Longitudinal changes, Fat mass index

INTRODUCTION

The increasing prevalence and negative consequences of obesity are demanding more valid and feasible field techniques to measure fat mass in populations at risk(1). Children with obesity present a higher risk of having cardiovascular diseases, diabetes, cancer and premature death later in life(2,3) as well as a higher risk of having a disability pension(4). In this context, feasible (i.e., affordable) and simple but accurate assessment methods of body composition are essential to early detect obesity at first ages and to control its changes throughout childhood(5,6).

The most common methods for clinical and field-based setting assessments are the anthropometric ones, such as weight and height to estimate body mass index (BMI), body circumference or skinfold thickness (SKF) measurement(6,7). The Slaughter's Equations (Slg-Eq)(8) are the most reliable and valid for estimating body fat from SKF in pediatric population(9,10). Another level of common methods but more complex includes bioelectrical impedance analysis (BIA)(11,12) and dual-energy X-ray absorptiometry (DXA), being the latter widely used as a criterion method(6,7,13).

Few studies included the validity of both the Slg-Eq and BIA methods against DXA for estimating body fat percentage (BFP) in young population(14–18), with just two focused on children with overweight/obesity(14,18). To our knowledge, no longitudinal studies have compared changes over the time in body composition measurements including the three abovementioned methods altogether. Separately, there were some studies that evaluated BIA against DXA in children with overweight/obesity and adolescents(19–23) and we found only one study including other SKF-based equations against DXA in 38 children and adolescents with obesity(24). Overall, these studies concluded having several limitations and poor accuracy, although these methods seemed to be valid to monitor the direction of the change(21) or to consider the evaluation at group level rather than individually in BIA compared to DXA(20).

Despite the fact that the existing body of evidence is informative, a number of novel research questions need to be further addressed: 1) The validity of Slg-Eq and BIA against DXA in different degrees of obesity is clinically relevant to know whether these methods can be used in young individuals suffering severe and morbid obesity, as the phenotype at highest risk. In this regard, the available information is scarce. Two studies compared BIA to DXA in children and/or adolescents with obesity(22,23,25) and one study compared Slg-Eq to DXA(26). 2) Although it has been widely used, BFP has not been suggested as a good measure for overall adiposity and to predict health-related outcomes(27,28). Instead, using fat mass adjusted for height could be a better approach(28,29). The proposed Fat Mass Index (FMI) is less reported than other parameters in the literature(30), and there are few validation studies including height-adjusted measures in children and/or adolescents(28,31). 3) There is little information about how accurate Slg-Eq(24) and BIA(19–22) are in measuring changes in BFP and FMI over the time when compared to DXA as a criterion method.

The present study aimed: i) to examine the criterion validity of Slg-Eq and BIA for estimating BFP and FMI in different degrees of obesity (overweight, mild obesity and severe-morbid obesity), pubertal stages and sex in children, compared to DXA as the criterion method; and ii) to determine the degree of agreement between methods when measuring longitudinally the adiposity changes after a physical-exercise intervention program.

METHODS

Design and participants

The data presented in this study are part of the ActiveBrains project (http://profith.ugr.es/activebrains), which is a randomized controlled trial that examined the effects of a 20-week physical exercise program on different health outcomes in children with overweight/obesity. Children were recruited from the Pediatric Unit of the "San Cecilio" and "Virgen de las Nieves" University Hospitals of Granada (Spain). Full description and detailed information of the ActiveBrains project is available elsewhere(32). A total of 92 children with overweight/obesity aged 8-12 years (10.0±1.2 years; 34.8% girls) were included in this study performed from November 2014 to June 2016. Sample size and power estimations were computed using IBM-SPSS Sample power software (version 3.0.1) for the intervention study; the same sample size has been used in all the ActiveBrains cross-sectional studies. See the ActiveBrains protocol study for more details in sample calculations(32).

Parents or legal guardians were informed of the purpose of the study and written informed parental consents were obtained. No remuneration was offered and the incentive was to partake in an advanced and controlled exercise program supervised by Physical Education Specialists for free. The ActiveBrains project was approved by the Ethics Committee on Human Research (CEIH) of the University of Granada (Reference: 848, February 2014), and registered in the ClinicalTrials.gov (Identifier: NCT02295072).

Measurements

Anthropometry

Anthropometric measurements were performed by the same experienced researcher following standardized techniques. The instruments were calibrated prior to use and all measurements were taken on the participant's left side according to specific protocols for children and adolescents(33,34).

Body weight was measured with an electronic scale (SECA 861, Hamburg, Germany) to the nearest 0.1 kg, height with a stadiometer (SECA 225, Hamburg, Germany) to the nearest 0.1 cm (children were instructed to take and hold a deep breath while the evaluator kept the child's head in the Frankfurt plane). All measurements were performed twice not consecutively with participants having bare feet and wearing underclothes, and averages were calculated.

SKF thicknesses (triceps and subscapular) were taken using a calliper (Holtain Ltd, UK) to the nearest 0.2 mm, they were measured twice not consecutively and, in order to ensure an accuracy of the measurement when the measures differed more than 2 mm, a third measure was taken and the average was used in further calculations. Briefly, Triceps SKF was measured halfway between the acromion process and the olecranon process, and the subscapular SKF about 20mm below the tip of the scapula, 45º to the lateral side of the body.

Body Composition

BMI was calculated as body mass divided by stature squared and expressed as kg/m^2 and it was categorized into three weight status, *i.e.*, overweight, obesity type I and obesity type II-III, according to the international standard cut-offs by age and gender(35–37). Since only 10 children were classified in obesity type III, they were grouped into obesity type II-III category for the analyses.

BFP and FMI calculated as body fat mass (FM) divided by stature squared and expressed as kg/m^2 were estimated using three different methods: i) Slg-Eq(8) from SKF thicknesses (triceps and subscapular). ii) BIA (manufacturer's equations), TANITA single frequency (50 KHz) bioimpedance scale (BC-418 MA, TANITA International Division, TANITA, UK); range: 2–200 kg; precision: 0.1 Kg; BFP range: 1%– 75%; BFP increments: 0.1%) was used to estimate the BFP, after a single measure. Participants were measured in a fasting state and according to manufacturer's instructions. And iii) DXA, (Discovery DXA system from Hologic, Marlborough, Massachusetts, USA), using the software version APEX 4.0.2 (Hologic Series Discovery QDR, Bedford, MA, USA). DXA equipment was calibrated using a lumbar spine phantom as recommended by the manufacturer. Participants' whole body was scanned in a supine position by the same researcher following standardized protocols(38) and specific recommendations for the analyses(39). DXA was used as a criterion method to test the validity of Slg-Eq and BIA.

Biological maturation

Pubertal stage development was assessed by Pediatricians using the scale by Tanner and Whitehouse(40), according to the breast and pubic hair development in girls and genital and pubic hair development in boys. Tanner's method is based on a scale of physical development in children, adolescents and adults. This scale defines the degree of maturational status according to sexual characteristics and groups them into 5 different stages (Tanner I-V). There were only 4 children classified into pubertal stage IV so they were grouped into the stage III category for the analyses.

Statistical analysis

All statistical analyses were performed using IBM SPSS Statistic software version 20.0 for Windows (SPSS Inc., Chicago, IL, USA). The level of significance was set at <0.05 for all the analyses. Descriptive data of the study sample are presented as percentages, means and standard deviations (SD). The accuracy and degree of agreement using Slg-Eq and BIA compared to DXA (in pre- and post-intervention) and the inter-method agreement between pre- post-intervention changes were assessed by calculating the bias or systematic error (mean difference) and random error as indicated by SD and 95% of Limits Of Agreement (LOA=1.96*SD) of the differences in BFP and FMI. Furthermore, we calculated the mean absolute percent error (MAPE) between the BFP and FMI estimates from Slg-Eq and BIA and compared them to those from DXA. Bland-Altman plots were depicted to study agreement of Slg-Eq and BIA with DXA measures of BFP and FMI.

Differences in the inter-method agreement by weight status, pubertal stage and sex were examined by one-way ANOVA (Analysis of Variance), entering sex, pubertal status or weight status as fixed factors in different models and the inter-method difference in body composition markers as dependent variables.

RESULTS

The characteristics of the participants (n=92) are shown in Table 1 and Supplementary Table 1. Strong to very strong partial (controlled by pubertal stage) correlation coefficients between methods were found for BFP (r range from 0.66 to 0.88), FM (r range from 0.89 to 0.97) and FMI (r range from 0.86 to 0.97); (All $P<0.001$), data not shown. Pre- and post-intervention bias (*i.e.*, systematic error) between Slg-Eq and BIA methods compared to DXA can be seen in Table 2 and Supplementary Table 2. Both Slg-Eq and BIA underestimated BFP, FM and FMI compared to DXA, being markedly smaller for Slg-Eq than BIA: *e.g.*, PRE showed BFP: -1.7% for Slg-Eq; and -7.6% for BIA and POST showed BFP: -2.3% for Slg-Eq; and -8.5% for BIA. On the other hand, the average SD (*i.e.*, random error) from pre- and post-intervention BFP assessments was larger for Slg-Eq (around 5.6 SD) than for BIA (around 3.0 SD). In regard to sex differences, Slg-Eq's underestimation of BFP, FM and FMI compared to DXA was larger in girls than in boys (all $p \leq 0.002$), being the bias in boys nearly 0.0. BIA showed no significant differences by sex (all $p > 0.700$). Agreement between methods can be visually observed in Bland-Altman plots for pre- and post-intervention time points (Figure 1 and Supplementary Figure 1). Slg-Eq show a positive trend from underestimation to overestimation compared to DXA as the fat indicators increase in participants, while the BIA estimates show no trends and closer LOA than the Slg-Eq. The results were highly consistent for the two assessment time points, i.e. pre- and post-intervention.

Figure 2 shows inter-method pre-intervention differences (Slg-Eq against DXA; FMI against DXA) in BFP and FMI across weight status (*i.e.*, overweight, obesity type I, obesity type II-III) and pubertal stage. The underestimation of BFP and FMI when comparing either Slg-Eq or BIA against DXA became generally smaller as the weight status increased. Although there were no significant differences by pubertal stage (all $p > 0.200$), the underestimation of BFP and FMI from Slg-Eq method seemed to be larger in less mature participants (*i.e.*, Pubertal Stage I *vs.* II and III) and the biases in FMI seemed to be closer to 0.0 with lower random error than in BFP. Again, BIA showed a larger underestimation of BFP and FMI than Slg-Eq. The analysis for FM shows the same trend that it has been described for FMI (Supplementary Figure 2).

The BFP and FMI changes from pre- to post-intervention according to the three different methods are shown in Figure 3. Slg-Eq and DXA methods were closer than BIA in the estimation of all parameters in both pre- and post-intervention assessments, with BIA reporting the lowest levels of adiposity. BFP (pre-intervention means from -0.0 to -0.7%) and FMI (pre-intervention means from 0.07 to -0.16 Kg/m^2) showed a slight decrease after the intervention mainly when using Slg-Eq or BIA. Supplementary Figure 3 shows FM changes, where Slg-Eq and DXA were closer than BIA as it has been described for BFP and FMI, but all methods showed a slight FM increase after the intervention (from 0.49 Kg to 1.23 Kg).

Table 3 and Supplementary Table 3 show the inter-method agreement on pre- post-intervention changes assessed by Slg-Eq and BIA compared to DXA as the criterion method. Generally, changes from pre- to post-intervention in adiposity parameters detected by the three methods were rather similar, with the largest inter-method difference observed in BFP in boys (*i.e.*, -0.7% for the comparison between Slg-Eq and DXA; and 0.8% for the comparison between BIA and DXA). In line with the results presented above, the bias between Slg-Eq and DXA was slightly smaller than between BIA and DXA in BFP, FM and FMI, and BFP SDs were larger in Slg-Eq changes than for BIA.

DISCUSSION

The present study contributed to the existing literature with the following findings: 1) Both the Slg-Eq and BIA methods underestimated (compared to DXA) BFP and FMI. This underestimation was smaller in Slg-Eq than in BIA (i.e., systematic error), yet the individual inter-method variation was larger in Slg-Eq than in BIA (i.e., random error).

2) Sex-differences were observed in the inter-method agreement for Slg-Eq but not for BIA. 3) Overall, the underestimation of adiposity by both methods was smaller as the weight status increased. 4) Finally, changes in adiposity were similarly captured by the three methods used.

Cross-sectional study of the agreement between methods

Previous validation studies have reported low agreement in both Slg-Eq(9,24) and BIA(19,20) when compared to DXA. Nonetheless, Slg-Eq have been shown as a valid and reliable method to asses adiposity in children and adolescents(9,10,41), as well as BIA has been shown as an alternative and promising method to assess nutritional status and growth(11,15). Despite these controversial findings, these methods have been recommended as suitable methods to be used in case that more accurate and reliable methodologies are not available(26), either in field or clinical settings(9,11). Furthermore, some authors have suggested to combine SKF with BIA to afford a greater approach to detect adiposity(7).

In the present study, both Slg-Eq and BIA methods (BFP: around -2.0% for Slg-Eq; and around -8.0% for BIA) underestimated adiposity when compared to DXA as the criterion method. In line to our results, the study of Noradilah et al.(14) in children aged 7-11 years also found that both Slg-Eq (-9.5±3.1%) and BIA (-4.9±5.3%) underestimated the BFP baseline values estimated by DXA. Although BIA results may not be compared between studies since the equipment used in our study was different from that used in Noradilah's study, they found a lower SD for Slg-Eq than for BIA, whereas we obtained a lower mean bias with wider SD for Slg-Eq BFP value (-1.7±5.6%) in pre-intervention and higher bias with lower SD for BIA (-7.6±2.9%). A possible explanation of these wide SD differences for Slg-Eq could be the larger SKF thicknesses found in children with higher adiposity. In fact, in the present study we observed that this random error seemed to be higher in severe-morbid obesity than in overweight or type I obesity. Noradilah et al.(14) included though a complete range of weight status (i.e. from severe thin to obese), which might also explain the higher variability between measurements. This idea has also been reported in the recent study of González-Ruiz et al.(18) in children and adolescents (11-17y) with excess of adiposity. They also found that BIA and Slg-Eq underestimated BFP compared to DXA, and that the differences between Slg-Eq against DXA could be related to the larger SKF thicknesses, since Slaugther et al.(8) included a sample with lower adiposity to develop their equations. In our children (80% of pre- and 20% of pubescents), the mean sum of triceps and subscapular SKF was 52.1±10.5 mm for boys and 55.2±12.5 mm for girls, while the sum of the sample of Slaughter et al.(8) was 17.7±6.9 mm for boys and 22.0±5.3 mm for girls. González-Ruiz et al.(18) obtained a sum of 37.2±8.5 mm for girls, taking into account that the data shown were from post-pubescents. Other studies only focused on BIA against DXA(19,20) also underestimated the results of adiposity, what concurs with our findings.

Altogether, it might seem that these common and widely used methods mostly underestimate adiposity when they are compared to a criterion method (i.e. DXA). However, there are also studies showing opposite findings. For instance, Thivel et al.(22) and Verney et al.(23) found that BIA overestimated fat mass and BFP compared to DXA in adolescents with obesity.

The agreement between the methods when measuring fat in children/adolescents with different weight status or degrees of obesity is controversial. In our study the biases of Slg-Eq and BIA compared to DXA were lower as the adiposity levels increased, especially when Slg-Eq were used. In line with our findings, the study of Seo et al.(25)

found better agreement between BIA and DXA in the group of children and adolescents with severe obesity than the group with mild to moderate obesity. Opposite findings were found by Freedman et al.(26) in children and adolescents aged 5 to 18y, ranked by the sum of SKF and classified in 4 sum categories according to 33rd, 67th and 90th percentiles within each sex. In their study Slg-Eq were fairly accurate among children without obesity but overestimated BFP with respect to the DXA of the children with obesity showing higher SKF thicknesses: up to 12% in boys and 5.8% in girls from the groups of sum of SKF higher than 60mm and 50mm respectively(26). Also, the study of Verney et al.(23) focused on adolescents with obesity divided in tertiles showed that BIA accuracy level decreased with increasing obesity compared to DXA.

According to sex differences, we found that BFP and FMI from the Slg-Eq were significantly underestimated in girls compared to boys. BFP bias was around -0.5% in boys and around -4.7% in girls using the Slg-Eq method. Similar tendency was observed by González-Ruiz et al.(18), who found that Slg-Eq underestimated BFP 9.0% in boys and 11.1% in girls. We did not find significant differences by sex using BIA, but it seemed to be slightly the opposite tendency that González-Ruiz et al.(18) found, where mean bias was larger in boys (14.0%) than in girls (11.0%). We found no data about sex differences in FMI to compare with our results.

It is more common that studies analyze adiposity through BFP rather than using FMI. This fact is in line with Zanini et al.(30) who concluded in their review that FMI had been seldom reported and that height adjustment should be used to control the effect of different heights. The use of FMI is more recent and it seems to be a better option than BFP to predict mortality due to cardiovascular causes(29). In our study we included FMI but we did not find significant differences across weight status or pubertal stages (FMI bias estimated from Slg-Eq was nearly 0.0 Kg/m^2 in obesity type I and in pubertal stages II and III). Slg-Eq showed an estimation of FMI closer to DXA than BIA, that showed similar underestimation regardless of weight status and pubertal stage. The study of Lewitt et al.(42) found that BFP from Slg-Eq was dependent on height in early puberty (Tanner stages I-III), but not in the late puberty (Tanner stages VI-V). Previously, Wells et al.(43) also reported that FM estimated by doubly labelled water was influenced by height in 8 year old children.

Longitudinal study of the agreement between methods

An important contribution of the present study was to examine the degree of agreement between methods over the time, where the magnitude of the change detected was similar among the three methods, i.e. the systematic errors observed in the pre-intervention were maintained in the post-intervention. Studies validating the SKF or BIA methods for estimating changes in body composition are scarce. The only study we found including SKF in children and adolescents with obesity was the one by Watts et al.(24), in which no significant correlations between FM changes from Slg-Eq (triceps and calf equation) and FM changes from DXA were observed after 8 weeks of intervention, being the SKF methods included in the study poorly predictive to assess changes in body composition. Continuing with BIA, Kasvis et al.(21) found a good degree of agreement between BIA and DXA with respect to the direction of the FM change over the time in children and adolescents with overweight and obesity, aged 7 to 13 years. Another study by Lyra et al.(19) in children and adolescents with the same weight problems showed that BIA underestimated FM in relation to DXA, being DXA more sensitive to body composition changes (i.e. FM and FFM), while BIA only detected changes in FM. Wan et al.(20) found in adolescents with obesity that BIA underestimated about 4kg of FM against DXA, but the estimated change of BFP over the time was similar between the two methods with wide limits of agreement. Finally, the recent study of Thivel et al.(22) in adolescents with obesity found higher differences in BFP and FM between BIA and DXA as the degree of changes increased, showing a reduced reliability to track body composition changes. We found that change bias in adiposity measured using the Slg-Eq and BIA methods were similar, with a maximum difference of around -0.7% in BIA with respect to DXA. Slg-Eq again showed lower systematic error and higher random error than BIA when compared to DXA, especially in BFP, and change biases (either Slg-Eq or BIA) were slightly higher in boys than in girls with no significant differences in any of the parameters (BFP and FMI). This consistency supports Slg-Eq and BIA as useful methods when measuring the differences of the changes over the time compared to DXA in children with overweight and obesity.

Limitations and strengths

This study has some limitations and strengths that must be taken into account. Since the study was aimed to test validity of Slg-Eq and BIA in children with overweight and obesity, it is unknown the extent to which these results would apply to children with normal-weight, underweight, or other populations. Moreover, anthropometric measurements and BIA also have their limitations since standardized protocols and specific conditions are needed to reduce the possibility of bias. It is important to note that the evaluator doing the skinfold measurements is certified by the ISAK and has extensive experience. The results derived from skinfold measurements are therefore more dependent on the expertise of the evaluator than when using other methods more automatized such as BIA. Finally, BIA could be influenced by hydration status. In our study, we did not control participants' hydration, but they were asked to come in a fasting status (water intake was not restricted) and to abstain from vigorous exercise the previous day. Strengths of this study include that all the DXA scans, BIA and anthropometric measurements were performed by the same trained evaluators, reducing therefore the inter-evaluator errors. In addition, the study of the capacity of Slg-Eq and BIA to detect changes in adiposity compared to DXA is a major strength of this study. Likewise, the study of how the measurement agreement might differ by degrees of obesity, maturational status, sex, and the inclusion of FMI, a newer and promising adiposity marker that should be acknowledged.

CONCLUSION

In conclusion, at a group level, Slg-Eq provided a relatively close estimation of adiposity (i.e. -2% of BFP) compared with DXA in children with overweight/obesity, yet it shows a substantial individual variation. On the other hand, BIA showed a large underestimation (-8% of BFP) compared to DXA, but had a lower individual variation and no inter-method differences between boys and girls. Despite these differences in the agreement with DXA at one time point (i.e., pre-intervention), both Slg-Eq and BIA methods agreed well in the direction and roughly in the magnitude of the change with DXA. Collectively, these findings support the notion that there might be an important estimation error when using Slg-Eq or BIA in comparison to DXA in children with overweight/obesity, but they can be considered a good feasible alternative to monitor changes in adiposity over the time.

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Table 1. Baseline characteristics of the participants

	Total			Boys			Girls		
	N	(%)		N	(%)		N	(%)	
Sex	92			60	(65.2)		32	(34.8)	
Weight Status*	88			57			31		
Overweight	21	(23.9)		14	(24.6)		7	(22.6)	
Obesity type I	38	(43.2)		26	(45.6)		12	(38.7)	
Obesity type II-III	29	(33.0)		17	(29.8)		12	(38.7)	
Pubertal Stage†	90			59			31		
Stage I	34	(37.8)		21	(35.6)		13	(41.9)	
Stage II	38	(42.2)		29	(49.2)		9	(29.0)	
Stage III	18	(20.0)		9	(15.3)		9	(29.0)	
	N	Mean	±SD	N	Mean	±SD	N	Mean	±SD
Age (years)	88	10.0	±1.2	57	10.2	±1.2	31	9.7	±1.1
Weight (Kg)	88	56.3	±11.3	57	56.8	±11.3	31	55.3	±11.3
Height (cm)	88	144.4	±8.6	57	145.0	±8.2	31	143.4	±9.3
BMI (Kg/m^2)	88	26.8	±3.5	57	26.8	±3.5	31	26.7	±3.5
Sum of 2 Skinfolds (mm) ‡	88	53.2	±11.3	57	52.1	±10.5	31	55.2	±12.5
Body Fat Percentage (%)									
Slg-Eq	88	41.5	±7.8	57	42.4	±8.1	31	39.8	±6.9
BIA	85	35.6	±5.9	55	34.9	±5.8	30	36.8	±6.1
DXA	89	43.1	±4.5	58	42.4	±4.1	31	44.4	±5.0
Fat Mass Index (Kg/m^2)									
Slg-Eq	88	11.3	±3.4	57	11.6	±3.5	31	10.8	±3.1
BIA	85	9.5	±2.7	55	9.3	±2.7	30	9.8	±2.7
DXA	87	11.4	±2.5	56	11.3	±2.4	31	11.8	±2.7

* Weight Status classification according to International Obesity Task Force (IOTF)(35–37)
† Pubertal Stage according to Tanner and Whitehouse(40)
‡ Sum of triceps and subscapular skinfolds

Table 2. Pre- and post-intervention (cross-sectional analysis) bias (mean differences±SD) and 95% limits of agreement between the Slaughter's Equations (Slg-Eq) and Bioelectrical Impedance Analysis (BIA) methods compared to Dual-energy X-ray Absorptiometry (DXA) as the criterion method.

	Total					**Boys**					**Girls**					
	N	Bias (mean diff.)	SD	95% limits of Agreement*	MAPE	N	Bias (mean diff.)	SD	95% limits of Agreement*	MAPE	N	Bias (mean diff.)	SD	95% limits of Agreement*	MAPE	P_{sex}‡
PRE-INTERVENTION																
Body Fat Percentage †																
Slg-Eq minus DXA	87	-1.69	±5.60	(-12.67; 9.29)	11.1%	56	-0.12	±5.73	(-11.35; 11.11)	10.7%	31	-4.53	±4.09	(-12.55; 3.49)	11.8%	P<0.001
BIA minus DXA	85	-7.56	±2.89	(-13.22; -1.90)	17.9%	55	-7.56	±2.92	(-13.28; -1.84)	18.2%	30	-7.55	±2.90	(-13.23; -1.87)	17.3%	P=0.982
Fat Mass Index (Kg/m²)																
Slg-Eq minus DXA	87	-0.16	±1.52	(-3.14; 2.82)	10.61	56	0.27	±1.61	(-2.89; 3.43)	10.6%	31	-0.93	±0.96	(-2.81; 0.95)	10.6%	P<0.001
BIA minus DXA	85	-2.00	±0.69	(-3.35; -0.65)	18.5%	55	-1.99	±0.71	(-3.38; 0.60)	18.8%	30	-2.02	±0.67	(-3.33; -0.71)	17.9%	P=0.834
POST-INTERVENTION																
Body Fat Percentage †																
Slg-Eq minus DXA	80	-2.26	±5.52	(-13.08; 8.56)	11.5%	52	-0.85	±5.30	(-11.24; 9.54)	10.6%	28	-4.90	±5.02	(-14.74; 4.94)	13.0%	P≤0.001
BIA minus DXA	63	-8.45	±3.16	(-14.64; -2.26)	19.8%	42	-8.52	±3.23	(-14.85; -2.19)	20.2%	21	-8.32	±3.06	(-14.32; -2.32)	18.8%	P=0.812
Fat Mass Index (Kg/m²)																
Slg-Eq minus DXA	80	-0.34	±1.48	(-3.24; 2.56)	11.1%	52	-0.05	±1.44	(-2.87; 2.77)	10.5%	28	-1.06	±1.30	(-3.61; 1.49)	12.0%	P≤0.001
BIA minus DXA	63	-2.26	±0.79	(-3.81; -0.71)	20.5%	42	-2.26	±0.83	(-3.89; -0.63)	20.9%	21	-2.27	±0.70	(-3.64; -0.90)	19.6%	P=0.983

SD, Standard Deviation; MAPE, Mean Absolute Percentage Error
* 95% limits of Agreement = SD*1.96
† Body Fat Percentage estimations from skinfolds were done according to Slaughter's equations(8)
‡ ANOVA was used to compare bias between boys and girls. Significant level was set at P<0.05

Table 3. Inter-methods agreement on pre- post-intervention change (longitudinal analysis) bias (mean differences±SD) and 95% limits of agreement as assessed by the Slaughter's Equations (Slg-Eq) and Bioelectrical Impedance Analysis (BIA) methods compared to Dual-energy X-ray Absorptiometry (DXA) as the criterion method.

	Total				**Boys**				**Girls**				
	N	Bias (mean diff.)	SD	95% limits of Agreement*	N	Bias (mean diff.)	SD	95% limits of Agreement*	N	Bias (mean diff.)	SD	95% limits of Agreement*	P_{sex}‡
Changes in Body Fat Percentage†													
Slg-Eq minus DXA	80	-0.60	±3.93	(-8.30; 7.10)	52	-0.73	±3.89	(-8.35; 6.89)	28	-0.36	±4.07	(-8.34; 7.62)	P=0.684
BIA minus DXA	61	-0.73	±2.62	(-5.87; 4.41)	41	-0.84	±2.59	(-5.92; 4.24)	20	-0.51	±2.74	(-5.88; 4.86)	P=0.647
Changes in Fat Mass Index (Kg/m^2)													
Slg-Eq minus DXA	80	-0.20	±1.13	(-2.41; 2.01)	52	-0.23	±1.13	(-2.44; 1.98)	28	-0.13	±1.15	(-2.38; 2.12)	P=0.686
BIA minus DXA	61	-0.23	±0.74	(-1.68; 1.22)	41	-0.26	±0.76	(-1.75; 1.23)	20	-0.18	±0.72	(-1.59; 1.23)	P=0.685

SD, Standard Deviation

* 95% limits of Agreement = SD*1.96

† Body Fat Percentage estimations from skinfolds were done according to Slaughter's equations(8)

‡ ANOVA was used to compare bias between boys and girls. Significant level was set at $P<0.05$

FIGURE TITLES AND LEGENDS

Figure 1. Bland-Altman plots for the pre- and post-intervention agreement of the Slaughter's Equations (Slg-Eq)(8) [Panel A] and Bioelectrical Impedance Analysis (BIA) [Panel B] with Dual-energy X-ray Absorptiometry (DXA) in the measurement of Body Fat Percentage (BFP) and Fat Mass Index (FMI).

Grey solid line represents perfect agreement (i.e., no difference).

Black dashed lines represent mean difference and limits of agreement between methods.

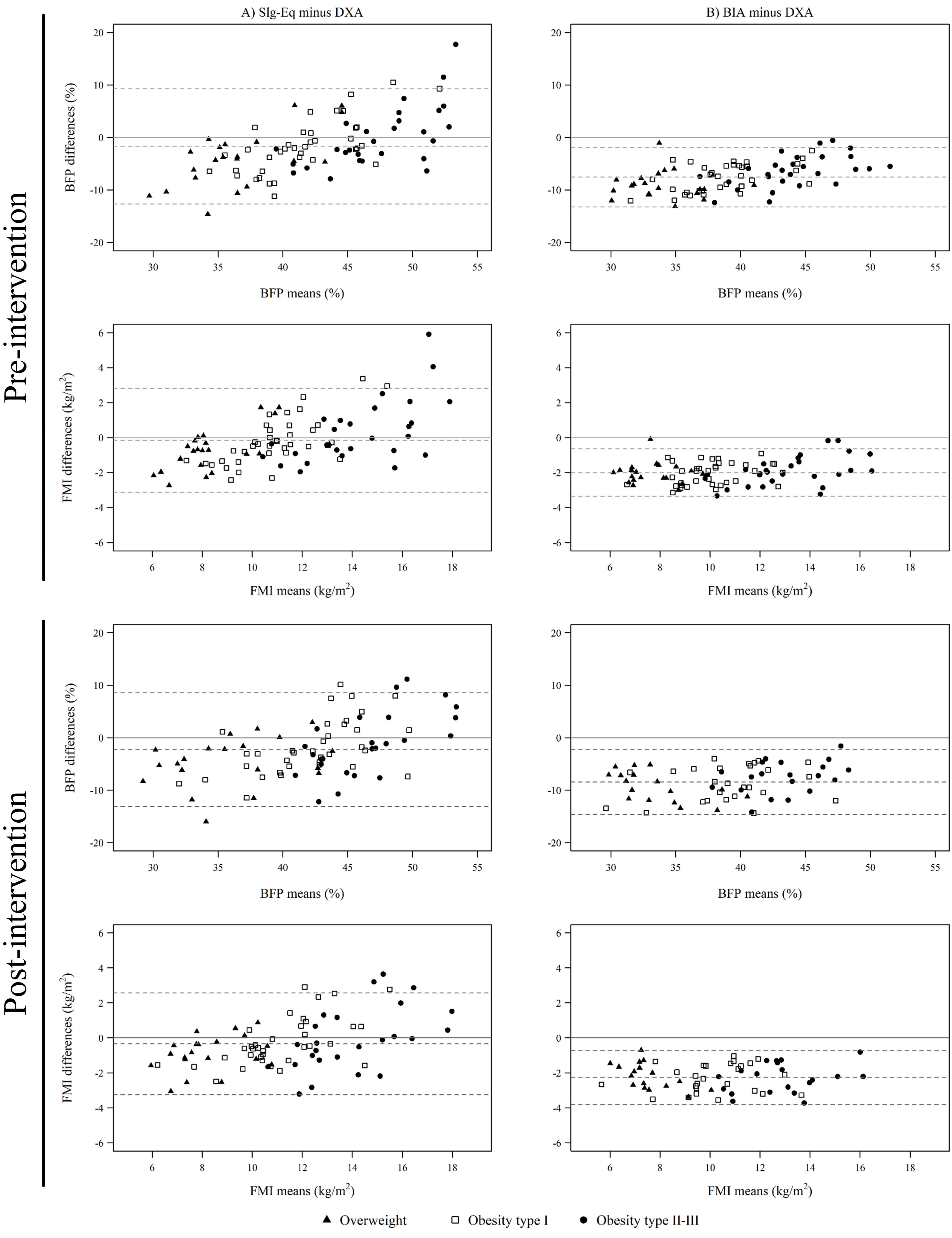
Figure 2. Inter-method pre-intervention differences (mean and CI 95%) in Body Fat Percentage (BFP) and Fat Mass Index (FMI) between the Slaughter's Equations (Slg-Eq)(8) and Bioelectrical Impedance Analysis (BIA) methods compared to Dual-energy X-ray Absorptiometry (DXA) as the criterion method across weight status (OW, Overweight; OB I, Obesity type I; OB II-III, Obesity type II and III) and pubertal stage.

Weight Status classification was done according to International Obesity Task Force (IOTF)(35–37)

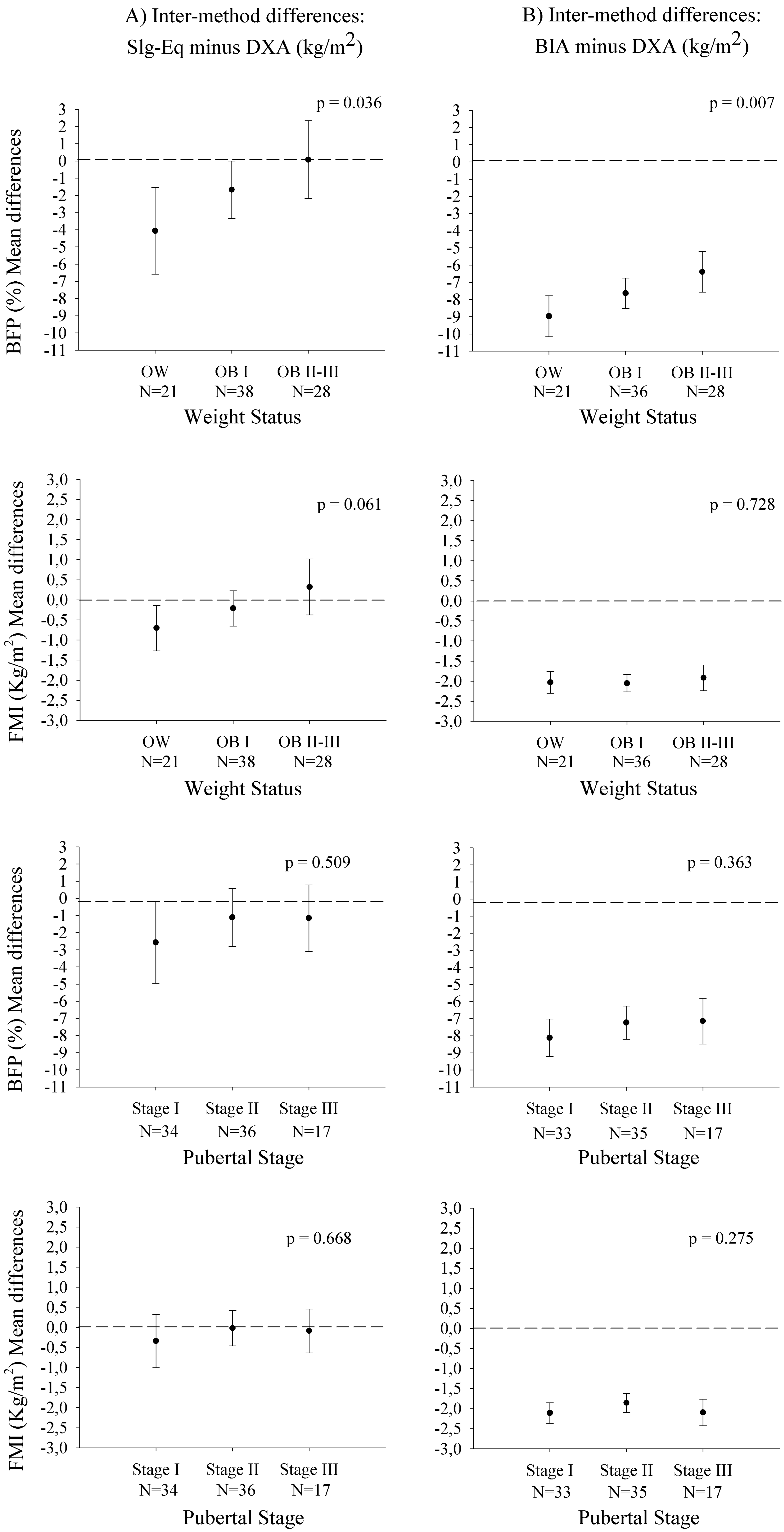
Pubertal stage according to Tanner and Whitehouse(40)

Figure 3. Descriptive intervention changes in Body Fat Percentage (BFP) and Fat Mass Index (FMI) estimated by Skinfolds using Slaughter's equations (Slg-Eq)(8), Bioelectrical Impedance Analysis (BIA) and Dual-energy X-ray absorptiometry (DXA).

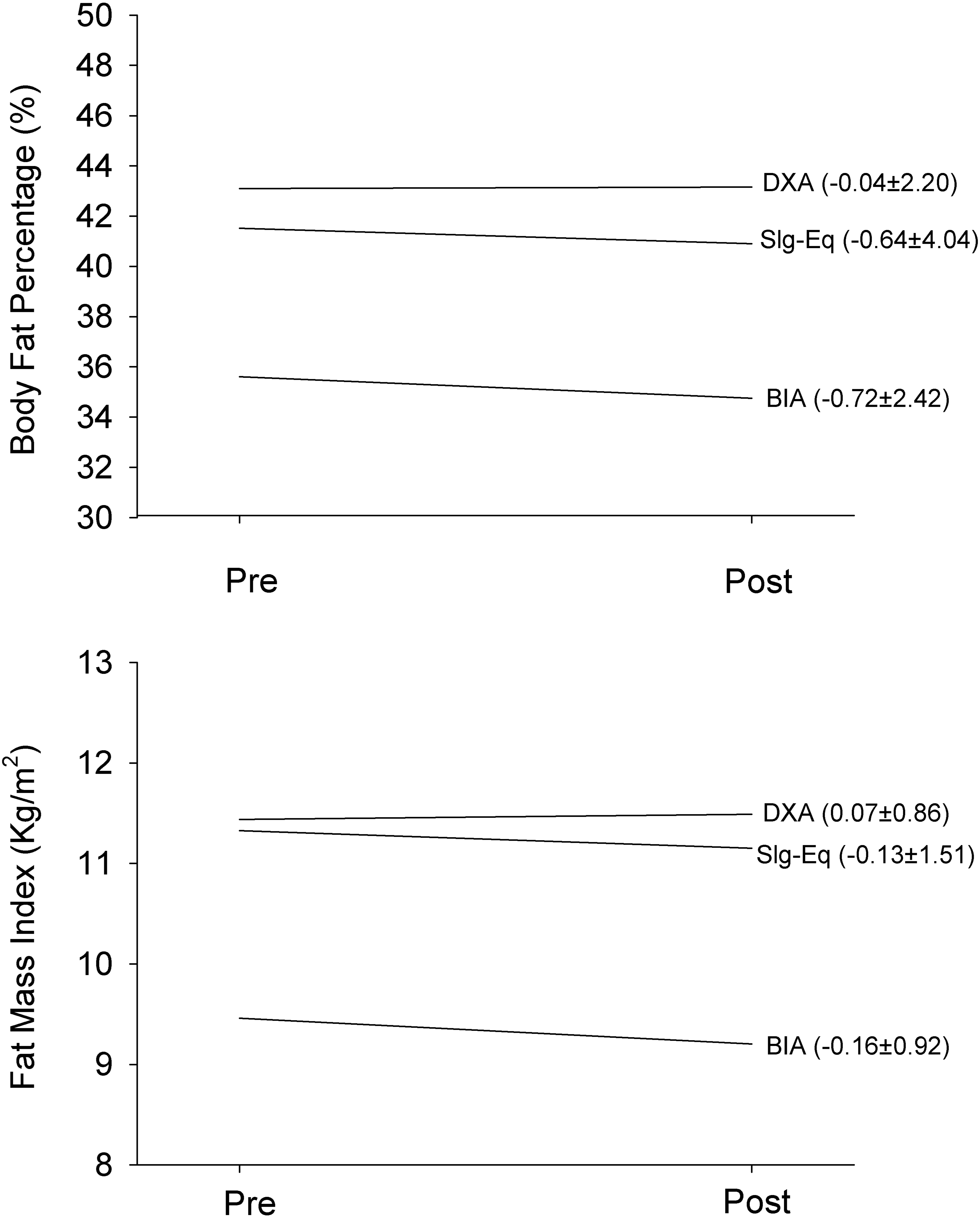
Data in parentheses are mean difference±SD of the change from pre- to post-intervention.



A) SIg-Eq minus DXA
B) BIA minus DXA
Pre-intervention
Post-intervention
BFP differences (%)
BFP means (%)
FMI differences (kg/m²)
FMI means (kg/m²)
Overweight
Obesity type I
Obesity type II-III



A) Inter-method differences:
Slg-Eq minus DXA (kg/m2)
B) Inter-method differences:
BIA minus DXA (kg/m2)
BFP (%) Mean differences
FMI (Kg/m2) Mean differences
p = 0.036
p = 0.007
p = 0.061
p = 0.728
p = 0.509
p = 0.363
p = 0.668
p = 0.275
OW N=21
OB I N=38
OB II-III N=28
OB I N=36
Weight Status
Stage I N=34
Stage II N=36
Stage III N=17
Stage I N=33
Stage II N=35
Pubertal Stage


Body Fat Percentage (%)
50
48
46
44
42
40
38
36
34
32
30
DXA (-0.04±2.20)
Slg-Eq (-0.64±4.04)
BIA (-0.72±2.42)
Pre
Post
Fat Mass Index (Kg/m2)
13
12
11
10
9
8
DXA (0.07±0.86)
Slg-Eq (-0.13±1.51)
BIA (-0.16±0.92)
Pre
Post