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Correlation of skin folds thickness and validation of prediction equations using DEXA as gold standard for estimation of body fat composition in Pakistani children

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ABSTRACT

Objective: to determine the correlation between Dual Energy X-ray Absorptiometry (DEXA) and skinfold thickness equations (SKF) for estimation of body fat composition in secondary school children and validation of prediction equations by Slaughter, Goran and Dezenberg.

Design: Cross sectional analytical study.

Setting: Joint Commission for International Accredited (JCIA) tertiary care hospital of Karachi, Pakistan from January 2010 till May 2010.

Participants: The study was approved by Hospital's ethical review committee. Written and verbal consents were obtained from principals of two schools and parents of 99 children (mean age of 14 ± 1.89 years; min-max 9-19 years; 54 male and 45 female) accrued in study. DEXA scan was acquired and skin fold thickness was measured at angle of the scapula, iliac crest and mid arm for bicep and triceps skin folds using Holtain calipers. Correlations were established between estimated fat mass (FM) and percentage body fat (%BF) calculated by DEXA and those predicted by prediction equations. On obtaining significant correlation of >0.5, overall accuracy and bias was calculated.

Results: There was an overall increased adiposity in females with fat mass of 3.57 kg and %BF 6.2% higher than male counterpart ($p < 0.05$). Slaughter equation predicted %BF accurately with overall high accuracy, minimal bias and with good agreement with DEXA. Dezenberg and Goran equations had significant bias in prediction of fat mass which was statistically significant with low level of accuracy of the Goran equation.

Conclusion: We conclude that Slaughter equation for estimating %BF showed strong validation with DEXA and can be used interchangeably in pediatric population. However, for estimation of fat mass, previously formulated equations by Goran and Dezenberg showed significant

difference in our population. We do recommend further studies for developing and validation of skin fold equations specific to Pakistani pediatric population.

STRENGTH AND LIMITATIONS: This is the first well designed and conducted study from a JCIA accredited tertiary care Hospital (Aga Khan University Hospital), Karachi Pakistan. In this study various well known equations for estimation of percentage body fat and fat mass in Western population have been validated for Pakistani children. Relatively smaller sample size is the basic limitation of this research output and warrants and provokes local researchers to do further well design studies in future.

KEY WORDS: Pediatric Obesity; DEXA; percentage body fat; fat mass; Slaughter; Goran; Dezenberg; skin fold thickness equations

INTRODUCTION

Obesity is now being reported as the new world epidemic afflicting more than1 billion adults globally[1]. According to WHO reports a great number of children (22 million) are afflicted with obesity with a great proportion in the United States, however the developing nations are not to be spared[2]. Analysis of the national health survey in Pakistan for adults reveals that lower BMI thresholds were suggested than those recommended in western population based on their association with outcome of chronic disease[3]. Same situation exists in children. Retrospective analysis of data in Britain revealed that there was an increased risk of obesity in Afro Caribbean and Pakistani girls whereas Indian and Pakistani boys were found to be at higher risk for overweight[4]. In another study it was reported that application of international standards did not provide a reliable result due to significant difference in height at the same ages of children belonging to the Asian and African descent as compared to standard population who showed a greater proportion of central obesity[5]. Based on these results emerged the concept of measuring body fat composition as standardization of BMI as an obesity measurement index across different ethnicities remains to be formulated.

Body fat composition is measured in children using dual energy x-ray absorptiometry (DEXA) as gold standard. Multiple validations of this technique with chemical composition, total body weighting and hydro densitometry have been performed[6-9]. Due to its very small amount of radiation and short procedure time it is ideal for use in children.[10]

However it is not suitable for use in field work as it is not widely available and involves transportation of participants to tertiary care settings. Another method that has been validated and widely used in the west is prediction equations based on anthropometric indices such as skinfold thickness of the biceps, triceps and abdominal wall as well as waist circumference and

waist hip ratios (Slaughter[11], Goran[12] and Dezenberg[13] equations). As availability of DEXA is limited to larger centers it is imperative to validate skinfold thickness equations for measurement of obesity in general population.

The purpose of this study was to determine the correlation between DEXA and skinfold thickness equations for estimation of body fat composition in secondary school children and validation of prediction equations (Slaughter, Goran and Dezenberg equations) against gold standard of DEXA by determining agreement and accuracy.

MATERIAL AND METHODS

This was a cross sectional analytical study conducted in Nuclear Medicine section, Department of Radiology, The Aga Khan University Hospital (AKUH), Karachi, Pakistan from Jan 2010 to May 2010-. The study was approved by AKUH ethical review committee.

The study population consisted of children enrolled form Class five till matriculate in the academic year from August of 2009 till May of 2010 form 2 community based schools belonging to the middle class strata. After the initial approval from the principal and obtaining school consent, a briefing lecture was delivered to the children with diagrammatic presentation of the machine and skin fold calipers. Thereafter assent forms to be signed by the children themselves and consent forms to be signed by the parents were distributed amongst the children. Furthermore phone numbers were obtained from the school administration and parents called to further explain the procedures and obtain verbal consent as well. All children whose parents gave consent were enrolled and those children from whom consent could not be obtained were excluded.

The sampling strategy was convenient sampling where consecutive children were enrolled till completion of sample size. A sample size of 44 subjects was needed to achieve 90% power to detect a change in slope from 0.0 under the null hypothesis to 0.45 under the alternative hypothesis when the standard deviation of the Triceps skin fold is 5.90, the standard deviation of Fat Mass by DEXA is 5.90, and the two-sided significance level is 0.05. These values were obtained from a study performed in children aged 4-10 years[14]. However since 2 schools were included, by considering a design effect of 2, the required sample size was calculated as 88 subjects.

Data collection was performed on a designed questionnaire for the study. It included the socio-demographic data of the subject and all independent variables. Training of the principal investigator (PI) and the data collector was done according to the National Health & Nutrition Examination Survey (NHANES) manual[15]. A senior radiologist trained the data collectors for identification of the anatomical surface landmarks where measurements of the skin fold were to be obtained namely the angle of the scapula, the iliac crest and the midpoint of the arm for measuring bicep and triceps skin folds. Intra and inter observer variability was assessed for 10 sets of reading and readings with a discrepancy for more than 0.4 mm were retaken and the procedure revised. Abdominal and hip circumference was measured using a measuring tape according to the described technique and a difference of more than one centimeter was considered unacceptable.

DEXA scan was performed on Hologic Discovery-A QDR series, USA. The parameters of the scan were alternating fan beam radiations of 140 kV and 100 kV with a scan width of 26.4 inches and scan length of maximum 77 inches (195 cm). The participant was asked to change in a hospital gown and asked to lie straight on the table with hands by the side palms facing down

away from the thighs and look at the ceiling to maintain head position. The body was positioned within the scan parameters marked on the table. The feet were rotated inwards so that the toes touched each other and taped so that the position was maintained. The method of performing the analysis of the estimation for %BF and total fat mass was done in the pediatric whole body scan mode for children up till 12 years of age. For children aged between 12 years onwards the adult scan mode was used for the calculation of body fat composition parameters. Reliability between scans was achieved with phantom calibration.

The measure of fat mass and %BF calculated by DEXA were received as a printout from the scanner and values were entered in the study performa. Prediction of the same parameters via preformed prediction equations (Slaughter's, Goran and Dezenberg) using skinfold thickness and anthropometric measures were also made.

Body measurements:

All body measurements were taken in accordance with the protocol described for anthropometric measurements in the National Health & Nutrition Examination Survey-3[15]. Body measurements were taken on the right side of the body.

Anthropometric measurements were taken using specially designed calipers (Holtain Calipers). The measurements included weight, triceps skinfold thickness and sub scapular skinfold thickness.

Weight was measured using a Tanita TBF 305 body fat analyser and weighing scales (Tanita UK Ltd, Yewsley, Middlesex, UK).

Measuring skin fold thickness:

Skinfolds measurements were taken to the nearest 0.1 millimeter. All skinfold measurements were done in duplicate (i.e., twice by the same investigator) since these measures have the most variability. Skinfold thickness was measured by Holtain calipers. The procedure was thoroughly explained to the children and the use of the caliper was demonstrated on the child's palm.

Data for all the independent variables was collected by the PI and assisting technician in the Dexa room. Two readings were taken for height, weight, abdominal and hip circumference, triceps, and subscapular and suprailiac skin fold thicknesses. If a difference of more than 0.3 mm was recorded the readings were retaken. Calibration of the calipers was performed periodically according to the calibration block provided by the manufacturers. The stadiometer and weighing scale were also calibrated and checked for correct position of zero before each data set recording.

Statistical analysis: Descriptive statistics were calculated for the data with mean and standard deviations for all continuous variables. Proportions were described for categorical data such as gender and independent T-test was applied to assess significant difference among gender.

The next step of the analysis was to validate the equation for fat mass and percentage body fat described by Goran, Dezenberg and Slaughter. In order to do this initially correlations were established between estimated FM and %BF calculated by DEXA and that predicted by using the equations. On obtaining significant correlation of >0.5, overall accuracy and bias was calculated.

Accuracy was defined as the degree of closeness of predicted fat mass and percent body fat obtained from the equations to their actual criterion value estimated by DEXA and would be considered significant if more than 90%. This was done by obtaining the overall mean of fat mass and %BF by DEXA as well as the Dezenberg, Goran and Slaughter equations. The

predicted mean of the equations was then subtracted from the mean value calculated by DEXA. The difference was then expressed as a percentage of the mean of the prediction equation.

Bias was defined as the systematic error within the data and was assessed as the average of the difference between FM and %BF measured by DEXA and that predicted by the equations and plotting the mean of errors against 0 using one sample t-test.

Agreement was defined as the degree of resemblance or consistency between the predicted and estimated values for fat mass and percent body fat. It was evaluated by applying the intra-class correlation coefficients.

Bland Altman plots were also generated for evaluation of agreement and bias. Based on the above mentioned criteria the equation for %BF by Slaughter and for fat mass by Dezenberg and Slaughter were rendered valid or rejected. A commercial Statistical Package for Social Sciences (SPSS Version 7) was used for these measurements.

RESULTS

Descriptive analysis and gender difference:

Ninety nine (n= 99) subjects were registered with a mean age of 14 ± 1.89 years (min-max 9-19 yrs) with 54 male and 45 female subjects. There was an evidence of overall increase in the adiposity of females as compared to males. All skin fold measures as well as fat composition variables were significantly higher in girls with p-value < 0.05. After assigning the binary variables it was observed that there was a difference of 3.57 kg fat mass present in excess in females as compared to the males as calculated by DEXA (p-value <0.0001). Significant differences were also obtained for %BF which was 6.2 % more in females as compared to males (p-value < 0.05). (Table 1)

After obtaining baseline data, the equations were applied to the sample for prediction of fat mass by Dezenberg and Goran as well as %BF by Slaughter. In the linear regression analysis strong correlations were obtained between the predicted values and the criterion values by DEXA with p-value<0.0001. (Table 2)

Validation of equations:

Overall accuracy:

The overall accuracy for the Slaughter equation was 98.4% which was very high and within the cutoff described previously. Dezenberg equation had an overall accuracy of 90.3% which was just within the cut-off designed. Goran equation showed weak overall accuracy of 83% which did not fall within the cutoff defined.

Calculating Bias:

No significant difference was obtained between the Slaughter equation predicting %BF and that estimated by DEXA with a bias of 0.37 (p-value 0.8). However, significant differences for predicting fat mass were obtained between DEXA and Dezenberg. The equation underestimated the fat mass with a bias of 1.26 kg which was statistically significant (p-value <0.0001). While Goran equation overestimated the fat mass with a bias of 1.48 kg which was statistically significant with a p-value of <0.0001. These results were further elaborated by the presentation of Bland Altman graphs in Figure 1.

Calculating intraclass correlation coefficients to assess internal validity:

Good correlations were identified ranging from 0.82-0.86 with significant p-values of <0.0001. This is suggestive of strong internal validity between the estimated and predicted values.

Thus in view of the above criteria the Slaughter equation fulfilled all the criteria for predicting %BF accurately as the overall accuracy is high with minimal bias which is not statistically significant and showing good agreement with %BF measured by DEXA. Furthermore more than 75% of the readings were within 5% of the DEXA values.

The Dezenberg and Goran equations had significant bias in prediction of fat mass which was statistically significant with low level of accuracy of the Goran equation. A summary of the results is shown in Table 3

DISCUSSION

The onset of obesity in the pediatric population has shown early precedence of atherosclerotic changes and diabetes mellitus with the onset of metabolic syndrome thus rendering obesity as a major health concern. Thus with this background we set out to conduct a study for the measurement of obesity in our pediatric population. A new technique of predicting body fat composition is by prediction equations (Salughter[11], Goran[12], and Dezenberg[13] equations) which have been developed for the western world. We attempted to validate these equations to predict body fat composition in our pediatric population which comprised of fat mass and %BF using DEXA as the gold standard. In this study only the Slaughter equation was validated for predicting %BF with a good correlation and high accuracy (98.4%). No significant bias was seen amongst the predicted and estimated method for measuring %BF with good intra class correlation coefficients (R = 0.82). These equations have been validated in different studies and used widely. In a study carried out by Janz in 122 subjects of pre and post pubescent children ranging from 8-17 years very high intra class correlations (0.98-0.99) and high validity correlations of (0.79-0.99) were achieved[16]. In another study conducted in Zaragoza, Spain carried out on 237 adolescents it was found that the Slaughter equations had the narrowest limits of agreement as compared to the other equations and did not show statistically significant bias[17]. Our results have been similar with the equation showing validity across all criteria.

Equations of fat mass by Goran and Dezenberg were not validated and showed significant bias. These results could be explained by the application of coefficients that were derived for the western population. Therefore determining coefficients for the predictors used in the Dezenberg equation and deriving a prediction equation that is sample specific could possibly yield better results. Another reason for inability to validate the Dezenberg equation might be that our sample

consisted of both pre and post pubescent children. The application of maturity specific coefficients by performing the Tanner staging might have different results as these were conducted on children who were prepubescent. Furthermore data regarding the ethnic origin and socio demographic status was also not applied. However all children were living in the same town going to community center schools with a middle class status. The Goran equation also was established on a cohort of children whose mean age was much lower than that of our sample. Furthermore gender was not accounted for in the equation we used whereas in our population significant differences were found between boys and girls for fat mass as well as all skin folds.

Very few studies have been done regionally in the South Asian population. In a study conducted in Sri Lanka in 282 children ages ranging from 5-15 years were tested for predicted %BF using multiple equations as well as Slaughter equation[18]. Although the bias calculated (-11.6 to 5.8) was not significant but the limits of agreement were high with significant underestimation of %BF rendering the equation invalid. An explanation for these results could be the criterion method that was used which was hydro densitometry. No results have been stated for validation against DEXA. In our study bias was not significant and more than 75% of the sample lied between a 5% variation as compared to DEXA. In 2010 a study was conducted in Hong Kong where children aged 9-19 were recruited and skin fold thickness were validated with subsequent formation of new equations[19]. However again the criterion used was air displacement plethysmography with only a subset validation with DEXA. They found significant difference between %BF estimated by the Slaughter equation although the absolute value was only of 1.52%. This was negated by our study which could be explained by the criterion method as being different and a significant difference in ethnicity. To the best of our understanding this is the first study conducted in Pakistani pediatric population to determine the correlation and validation

of skinfold thickness equations for estimation of body fat composition using DEXA as gold standard.

CONCLUSIONS

We conclude that the Slaughter equation for estimating %BF showed strong validation with DEXA and can be used interchangeably in pediatric population. However, for estimation of fat mass, the previously formulated equations by Goran and Dezenberg showed significant difference in our population. We do recommend further studies for developing and validation of skin fold equation specific to Pakistani pediatric population.

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CONTRIBUTORSHIP STATEMENT

All authors have substantially contributed to conception and design of study, acquisition of data, analysis and interpretation of data and preparatio of masnuscripty.

CONFLICT OF INTEREST:

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Table 1: Baseline Demographic and measured values of studied subjects

Total subjects: 99	Males = 54 mean (SD)	Females = 45 mean (SD)
Age (yrs)	13.8 (2.0)	14.13(2.2)
Weight (kg)*	41.7 (12.5)	43.6 (8.2)
BMI (kg/m^2) *	17.7 (3.5)	19.0 (3.4)
SC-SF (mm)*	9.9 (6.1)	14.1 (5.4)
T-SF (mm)*	11.4 (7.1)	16.0 (6.1)
FM by DZ-Eq	8.9 (5.4)	12.7 (4.1)
FM by G-Eq	20.7 (4.1)	28.6 (3.2)
%BF BY SL-Eq	10.7 (9.3)	13.3 (5.1)
DEXA FM*	8.3 (5.6)	10.2 (3.2)
DEXA %BF*	19.1 (7.6)	29.6 (5.1)

*: p value <0.05
BMI: Body Mass Index
SC-SF: Subscapular skinfold
T-SF: Tricep Skin Fold
FM: Fat Mass

%BF: Percentage Body Fat
DZ-Eq: Dezenberg Equation
G-Eq: Goran Equation
SL-Eq: Slaughter Equation

Table 2: Correlation coefficients calculated for prediction equations of DZ, G and SL for fat mass (FM) and percentage body fat (%BF). All equations showed good correlations with significant p-values of <0.0001.

Equations	R^2	SEE
• DZ-FM	0.8	2.45
• G-FM	0.86	2.06
• SL-% BF	0.76	3.73

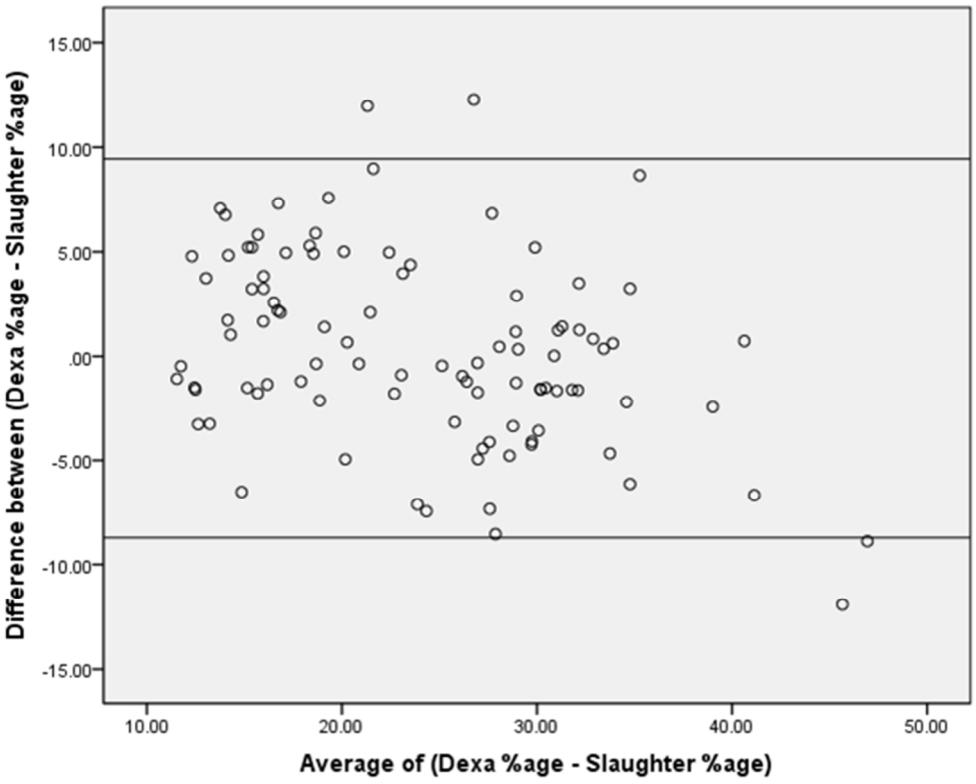
SEE: Standard Error of Estimate
DZ-FM: Dezenberg Fat Mass
G-FM: Goran Fat Mass
SL %BF: Slaughter Percentage Body Fat

Table 3: Validation criteria for prediction of % body fat and fat mass

	Slaughter	Goran	Dezenberg
Correlation	0.76	0.8	0.86
Accuracy	98.4	90.3	83.7
Bias	0.37 (p =0.4)	-1.26 (p<0.0001)	1.48 (p<0.0001)
Intra-class correlation	0.82	0.85	0.85

LEGENDS

Figure 1: (1a) Bland Altman plot shows uniform distribution between %BF calculated by DEXA and Slaughter. Although limits of agreement wide more than 75% of sample is between 5% variation; (1b) shows significant overestimation of fat mass between DEXA and Goran equation with almost the entire sample above the zero levels; (1c) shows significant underestimation of fat mass with more than 60% of the sample lying below the 0 level in the Dezenberg equation.



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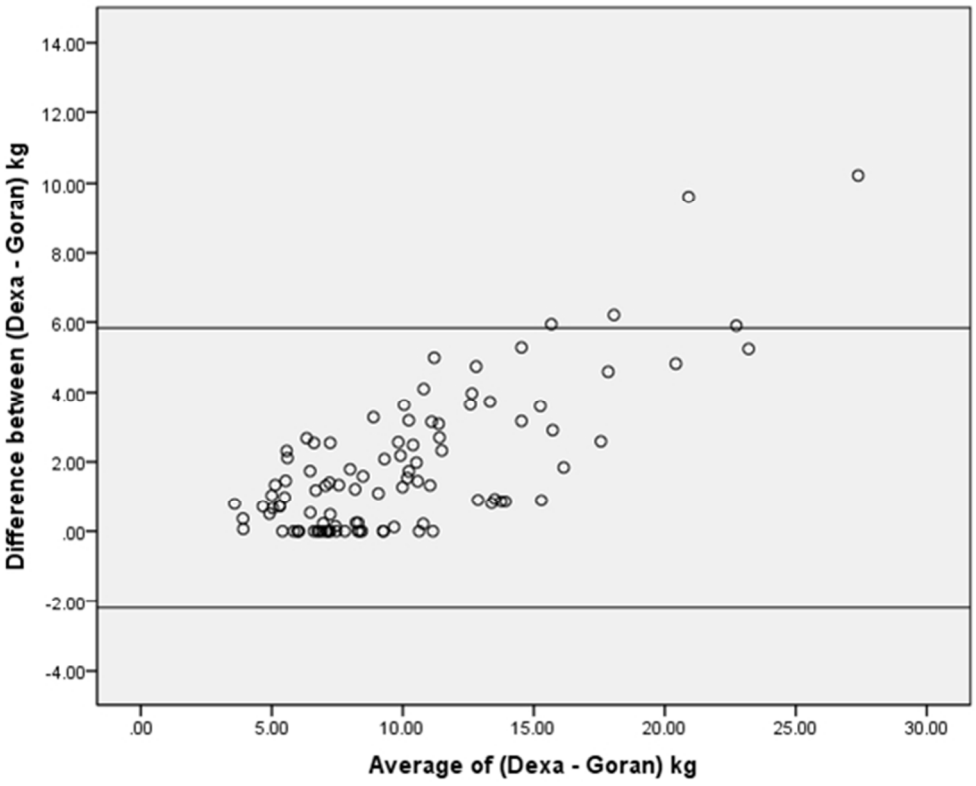

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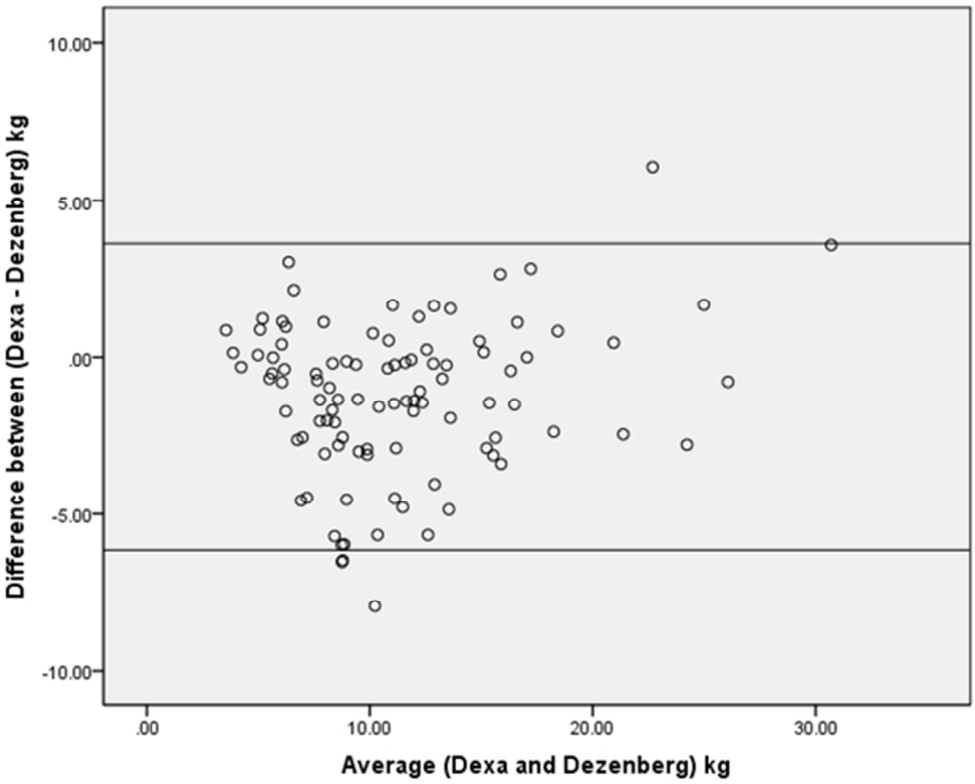

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ABSTRACT

Objective: to determine the correlation between Dual Energy X-ray Absorptiometry (DEXA) and skinfold thickness equations (SKF) for estimation of body fat composition in secondary school children and validation of prediction equations by Slaughter, Goran and Dezenberg.

Design: Cross sectional analytical study.

Setting: Joint Commission for International Accredited (JCIA) tertiary care hospital of Karachi, Pakistan from January 2010 till May 2010.

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Results: There was an overall increased adiposity in females with fat mass of 3.57 kg and %BF 6.2% higher than male counterpart ($p < 0.05$). Slaughter equation predicted %BF accurately with overall high accuracy, minimal bias and with good precision with DEXA. Dezenberg and Goran equations had significant bias in prediction of fat mass which was statistically significant with low level of accuracy of the Goran equation.

Conclusion: We conclude that Slaughter equation for estimating %BF showed reasonable validation with DEXA. Nevertheless further studies with consideration for maturity and ethnicity are warranted for better results. However, for estimation of fat mass, previously formulated

equations by Goran and Dezenberg showed significant difference in our population. We do recommend further studies for developing and validation of skin fold equations specific to Pakistani pediatric population.

STRENGTH AND LIMITATIONS: This is the first well designed and conducted study from a JCIA accredited tertiary care Hospital (Aga Khan University Hospital), Karachi Pakistan. In this study various well known equations for estimation of percentage body fat and fat mass in Western population have been validated for Pakistani children. Relatively smaller sample size is the basic limitation of this research output which warrants and provokes local researchers to do further studies in future.

KEY WORDS: Pediatric Obesity; DEXA; percentage body fat; fat mass; Slaughter; Goran; Dezenberg; skin fold thickness equations

INTRODUCTION

Obesity is now being reported as the new world epidemic afflicting more than1 billion adults globally[1]. According to WHO reports a great number of children (22 million) are afflicted with obesity with a great proportion in the United States, however the developing nations are not to be spared[2]. Analysis of the national health survey in Pakistan for adults reveals that lower BMI thresholds were suggested than those recommended in western population based on their association with outcome of chronic disease[3]. Same situation exists in children. Retrospective analysis of data in Britain revealed that there was an increased risk of obesity in Afro Caribbean and Pakistani girls whereas Indian and Pakistani boys were found to be at higher risk for overweight[4]. In another study it was reported that application of international standards did not provide a reliable result due to significant difference in height at the same ages of children belonging to the Asian and African descent as compared to standard population who showed a greater proportion of central obesity[5]. Based on these results emerged the concept of measuring body fat composition as standardization of BMI as an obesity measurement index across different ethnicities remains to be formulated.

Body fat composition is measured in children using dual energy x-ray absorptiometry (DEXA) as gold standard. Multiple validations of this technique with chemical composition, total body weighting and hydro densitometry have been performed[6-9]. Due to its very small amount of radiation and short procedure time it is ideal for use in children.[10]

However it is not suitable for use in field work as it is not widely available and involves transportation of participants to tertiary care settings. Another method that has been validated

and widely used in the west is prediction equations based on anthropometric indices such as skinfold thickness of the biceps, triceps and abdominal wall as well as waist circumference and waist hip ratios. The first equations that have been produced have initially derived by Slaughter et al [11]. In a sample of 310 children and adults age 8-29 yrs. prediction equations were cross validated against under body weighting using 4 compartment model, bone mineral studies and body water dilution. Special consideration for ethnicity and maturations was given. Multiple studies have tried to validate these equations in their population and on failing to do so derive their own equations. In a study by Goran et al they have previously cross validated DEXA against pig carcasses in the pediatric weight range and then after adjusting fat mass values with coefficient they have derived equations in prepubescent children by identifying the best predictors for fat mass and deploying them in a prediction equation[12].

As a second part of the Goran study, Dezenberg et al also tried to validate the Slaughter and Goran equations in two different cohorts (the Alabama and the Vermont cohort) across multiple criteria and on failure to do so went ahead and formulated their own equations by identifying best predictors [13]. They too also came up with the conclusion that although much work has been done larger cohorts with different ethnicities is needed to establish the authenticity of prediction equations for widespread use in field work. As availability of DEXA is limited to larger centers it is imperative to validate skinfold thickness equations for measurement of obesity in general population.

The purpose of this study was to determine the correlation between DEXA and skinfold thickness equations for estimation of body fat composition in secondary school children and validation of prediction equations (Slaughter, Goran and Dezenberg equations) against gold standard of DEXA by determining agreement and accuracy.

MATERIAL AND METHODS

This was a cross sectional analytical study conducted in Nuclear Medicine section, Department of Radiology, The Aga Khan University Hospital (AKUH), Karachi, Pakistan from Jan 2010 to May 2010-. The study was approved by AKUH ethical review committee.

The study population consisted of children enrolled form Class five till matriculate in the academic year from August of 2009 till May of 2010 form 2 community based schools belonging to the middle class strata. After the initial approval from the principal and obtaining school consent, a briefing lecture was delivered to the children with diagrammatic presentation of the machine and skin fold calipers. Thereafter assent forms to be signed by the children themselves and consent forms to be signed by the parents were distributed amongst the children. Furthermore phone numbers were obtained from the school administration and parents called to further explain the procedures and obtain verbal consent as well. All children whose parents gave consent were enrolled and those children from whom consent could not be obtained were excluded.

The sampling strategy was convenient sampling where consecutive children were enrolled till completion of sample size. A sample size of 44 subjects was needed to achieve 90% power to detect a change in slope from 0.0 under the null hypothesis to 0.45 under the alternative hypothesis when the standard deviation of the Triceps skin fold is 5.90, the standard deviation of Fat Mass by DEXA is 5.90, and the two-sided significance level is 0.05. These values were obtained from a study performed in children aged 4-10 years[14]. However since 2 schools were included, by considering a design effect of 2, the required sample size was calculated as 88 subjects.

Data collection was performed on a designed questionnaire for the study. It included the socio-demographic data of the subject and all independent variables. Training of the principal investigator (PI) and the data collector was done according to the National Health & Nutrition Examination Survey (NHANES) manual[15]. A senior radiologist trained the data collectors for identification of the anatomical surface landmarks where measurements of the skin fold were to be obtained namely the angle of the scapula, the iliac crest and the midpoint of the arm for measuring bicep and triceps skin folds. Intra and inter observer variability was assessed for 10 sets of reading and readings with a discrepancy for more than 0.4 mm were retaken and the procedure revised. Abdominal and hip circumference was measured using a measuring tape according to the described technique and a difference of more than one centimeter was considered unacceptable.

DEXA scan was performed on Hologic Discovery-A QDR series, USA. The parameters of the scan were alternating fan beam radiations of 140 kV and 100 kV with a scan width of 26.4 inches and scan length of maximum 77 inches (195 cm). The participant was asked to change in a hospital gown and asked to lie straight on the table with hands by the side palms facing down away from the thighs and look at the ceiling to maintain head position. The body was positioned within the scan parameters marked on the table. The feet were rotated inwards so that the toes touched each other and taped so that the position was maintained. The method of performing the analysis of the estimation for %BF and total fat mass was done in the pediatric whole body scan mode for children up till 12 years of age. For children aged between 12 years onwards the adult scan mode was used for the calculation of body fat composition parameters. Reliability between scans was achieved with phantom calibration.

The measure of fat mass and %BF calculated by DEXA were received as a printout from the scanner and values were entered in the study performa. Prediction of the same parameters via preformed prediction equations (Slaughter's, Goran and Dezenberg) using skinfold thickness and anthropometric measures were also made.

Body measurements:

All body measurements were taken in accordance with the protocol described for anthropometric measurements in the National Health & Nutrition Examination Survey-3[15]. Body measurements were taken on the right side of the body.

Anthropometric measurements were taken using specially designed calipers (Holtain Calipers). The measurements included weight, triceps skinfold thickness and sub scapular skinfold thickness.

Weight was measured using a Tanita TBF 305 body fat analyser and weighing scales (Tanita UK Ltd, Yewsley, Middlesex, UK).

Measuring skin fold thickness:

Skin fold measurements were obtained in various areas of the body such as the anterior and posterior aspect of the upper arm corresponding to the *Bicep* and *triceps* SF.

Sub scapular SF was obtained by measuring the subcutaneous tissue beneath the shoulder blade.

Suprailiac SF was obtained by measuring the fold above the pelvic bone laterally.

Abdominal SF was measured just adjacent to the umbilicus.

Abdominal circumference was measured using a measuring tape at the level of the umbilicus.

Hip circumference was measured at the maximum curvature of the hips.

Skinfolds measurements were taken to the nearest 0.1 millimeter. All skinfold measurements were done in duplicate (i.e., twice by the same investigator) since these measures have the most variability. Skinfold thickness was measured by Holtain calipers. The procedure was thoroughly explained to the children and the use of the caliper was demonstrated on the child's palm.

The following prediction equations were used:

Fat mass is obtained as amount of fat calculated in kilograms predicted by the DZ and G equations

Dezenberg equation:

FM (kilograms) = 0.342 × body mass (kg) + 0.256 × triceps SF (mm) + 0.837 × sex (1= boy, 2 = girl) - 7.388

Goran equation:

FM (kg) = 0.23 (sub scapular SF) + 0.18 (weight) + 0.13 (triceps SF) – 3.0

Percentage body fat is obtained as a percentage of body fat predicted by the Slaughter equation.

For boys:

%BF = 1.21 [sum of triceps and subscapular skinfolds in mm] - 0.008 [9sum of triceps and subscapular skinfolds in mm) ^{2}] - 1.7 (coefficient for white Prepubescent boys in boys with sum of triceps and subscapular skinfolds < 35 mm);

If sum of triceps and subscapular skinfolds > 35 mm,

%BF = 0.783[Sum of triceps and subscapular skinfolds mm] - 1.7 in boys;

For girls:

%BF = 1.33 [sum of triceps and subscapular skinfolds _mm] - 0.013 [(sum of triceps and subscapular skinfolds in mm) ^{2}] + 3.0 (coefficient for white girls) in girls with sum of triceps and subscapular skinfolds < 35 mm;

Or if sum of triceps and subscapular skinfolds > 35 mm,

%BF = 0.546 [sum of triceps and subscapular skinfolds in mm] + 9.7 in girls

Data for all the independent variables was collected by the PI and assisting technician in the Dexa room. Two readings were taken for height, weight, abdominal and hip circumference, triceps, and subscapular and suprailiac skin fold thicknesses and the average measurement for both the readings was taken as the final reading. If a difference of more than 0.3 mm was recorded the readings were retaken. Calibration of the calipers was performed periodically according to the calibration block provided by the manufacturers. The stadiometer and weighing scale were also calibrated and checked for correct position of zero before each data set recording.

Statistical analysis: Descriptive statistics were calculated for the data with mean and standard deviations for all continuous variables. Proportions were described for categorical data such as gender and independent T-test was applied to assess significant difference among gender.

The next step of the analysis was to validate the equation for fat mass and percentage body fat described by Goran, Dezenberg and Slaughter. In order to do this initially correlations were established between estimated FM and %BF calculated by DEXA and that predicted by using the 3 equations. On obtaining significant correlation of > 0.05, precision and bias was calculated.

Accuracy was defined as the degree of closeness of predicted fat mass and percent body fat obtained from the equations to their actual criterion value estimated by DEXA and would be considered significant if more than 90%. This was done by obtaining the overall mean of fat mass and %BF by DEXA as well as the Dezenberg, Goran and Slaughter equations separately. Individual accuracies were then calculated by subtracting the predicted mean of percent body fat and fat mass obtained from the equations from the mean value of fat mass and percentage body fat calculated by DEXA. The difference was then expressed as a percentage of the mean of the estimated DEXA value for the 3 equations..

Precision was defined as the variation between the predicted and estimated values of %BF and FM within the data and was assessed as the average of the difference between FM and %BF measured by DEXA and that predicted by the equations and plotting the mean of errors against 0 using paired samples t-test.

Bland Altman plots were also generated for evaluation of agreement and bias. Based on the above mentioned criteria the equation for %BF by Slaughter and for fat mass by Dezenberg and Slaughter were rendered valid or rejected. Post hoc regression analysis was also performed with the line of identity against predicted versus estimated fat mass and percent body fat. A commercial Statistical Package for Social Sciences (SPSS Version 17) was used for these measurements.

RESULTS

Descriptive analysis and gender difference:

Ninety nine (n= 99) subjects were registered with a mean age of 14 ± 1.89 years (min-max 9-19 yrs) with 54 male and 45 female subjects. There was an evidence of overall increase in the adiposity of females as compared to males. All skin fold measures as well as fat composition variables were significantly higher in girls with p-value < 0.05. After assigning the binary variables it was observed that there was a difference of 3.57 kg fat mass present in excess in females as compared to the males as calculated by DEXA (p-value <0.0001). Significant differences were also obtained for %BF which was 6.2 % more in females as compared to males (p-value < 0.05). (Table 1)

After obtaining baseline data, the equations were applied to the sample for prediction of fat mass by Dezenberg and Goran as well as %BF by Slaughter. In the linear regression analysis strong correlations were obtained between the predicted values and the criterion values by DEXA with p-value<0.0001. (Table 2).

Validation of equations:

Overall accuracy:

The overall accuracy for the Slaughter equation was 98.4% which was very high and Dezenberg equation had an overall accuracy of 90.3% which was just within the cut-off designed. Goran equation showed weak overall accuracy of 83% which did not fall within the cutoff defined.

Variation/Precision of equations

No significant difference was obtained between the Slaughter equation predicting %BF and that estimated by DEXA with a difference of 0.37 kg (p-value 0.425). However, significant differences for predicting fat mass were obtained between DEXA and Dezenberg. The equation underestimated the fat mass with a bias of 1.26 kg which was statistically significant (p-value <0.0001). While Goran equation overestimated the fat mass with a bias of 1.48 kg which was statistically significant with a p-value of <0.0001

Calculating Bias:

Limits of agreement were evaluated using Bland Altman analysis. The slaughter equation revealed that more than 75% of the data was within the first standard deviation.

Thus in view of the above criteria the Slaughter equation fulfilled all the criteria for predicting %BF accurately as the overall accuracy is high with minimal variation which is not statistically significant and showing good precision with %BF measured by DEXA. Furthermore more than 75% of the readings were within 5% of the estimated DEXA values.

The Dezenberg and Goran equations had significant variation in prediction of fat mass which was statistically significant with low level of accuracy of the Goran equation. A summary of the results is shown in Table 3

Post hoc regression analysis revealed that both the Goran and Dezenberg equation did not reveal an intercept significantly different from zero. The Slaughter equation did not reveal an intercept significantly different from zero against estimated fat mass however the intercept was significantly different from zero when it was assessed against estimated percent body fat (Fig 1).

DISCUSSION

The onset of obesity in the pediatric population has shown early precedence of atherosclerotic changes and diabetes mellitus with the onset of metabolic syndrome thus rendering obesity as a major health concern. Thus with this background we set out to conduct a study for the measurement of obesity in our pediatric population. A new technique of predicting body fat composition is by prediction equations (Salughter[11], Goran[12], and Dezenberg[13] equations) which have been developed for the western world. We attempted to validate these equations to predict body fat composition in our pediatric population which comprised of fat mass and %BF using DEXA as the gold standard. In this study only the Slaughter equation was validated for predicting %BF with a good correlation and high accuracy (98.4%). No significant bias was seen amongst the predicted and estimated method for measuring %BF. These equations have been validated in different studies and used widely. In a study carried out by Janz in 122 subjects of pre and post pubescent children ranging from 8-17 years very high intra class correlations (0.98-0.99) and high validity correlations of (0.79-0.99) were achieved[16]. In another study conducted in Zaragoza, Spain carried out on 237 adolescents it was found that the Slaughter equations had the narrowest limits of agreement as compared to the other equations and did not show statistically significant bias[17]. Our results have been similar with the equation showing validity across all criteria.

In the study by Goran et al, after failing to validate the Slaughter equation they derived an equation for their own sample. It was reported that all skin folds showed higher correlations with fat mass rather than percent body fat so equation for the prediction of fat mass was generated.

Another method of calculating fat composition was also deployed in this study using bioelectrical impedance analysis. They found the variables of triceps, sub scapular and weight significantly correlating. The equation was further cross validated on 1/3rd of the sample according to the criteria of Lohman which consisted of assessing the difference between the predicted and estimated fat mass as well as minimal increase of the SEE. Only the R^2 was reported of 0.88.

This equation was then further cross validated in the study by Dezenberg. In his study after initially failing to validate the Goran and Slaughter equation, a prediction equation was formulated across a sample of different ethnic cohorts consisting of both black and white population. The predictors considered significant were weight; triceps skin fold, gender, ethnicity and abdominal skin fold. However since we were not evaluating ethnicity, the equation adopted in this study consisted only of weight, triceps skin fold and gender predictors. A significant point of concern in this study was the fact that regression analysis was performed between percentage body fat as predicted by Slaughter against fat mass as predicted by dexa. Since both variables were in different units the inability to validate the equations appeared inevitable. In our study adopting the approach of Dezenberg et al we achieved good correlation with the intercept not significantly different from zero with Slaughter equations predicting percent body fat and fat mass in kilograms estimated by dexa (fig 2d) however the regression analysis between predicted percent body fat and that estimated with dexa shows an intercept significantly different from zero. Despite this as multiple validation criteria were applied and the Slaughter equation maintained robustness across all of them it can be used with some considerations of maturity specificity and ethnicity in our population.

Our results were similar to the results by Eisenmann who evaluated a number of equations along with bioelectrical impedance analysis. In his study he found that the Slaughter equations consisting of triceps and subscapular skinfold were showing better limits of agreement and reduced bias there only one of the slaughter equations was adopted. Significant differences were assessed between the methods for evaluating body fat composition by DEXA and SF equations, bioelectrical impedance.

A few issues are worthy of mention here. These results could be explained by the application of coefficients that were derived for the western population. Therefore determining coefficients for the predictors used in the Dezenberg equation and deriving a prediction equation that is sample specific could possibly yield better results.

Another reason for inability to validate the DZ equation might be that our sample consisted of both pre and post pubescent children. The application of maturity specific coefficients by performing the Tanner staging might have different results as these were conducted on children who were prepubescent. Furthermore data regarding the ethnic origin and socio demographic status was also not applied. However all children were living in the same town going to community center schools with a middle class status.

The Goran equation also was established on a cohort of children whose mean age was much lower than that of our sample. Furthermore gender was not accounted for in the equation we used whereas in our population significant differences were found between boys and girls for fat mass as well as all skin folds.

Very few studies have been done regionally in the South Asian population. In a study conducted in Sri Lanka in 282 children ages ranging from 5-15 years were tested for predicted %BF using multiple equations as well as Slaughter equation[18]. Although the bias calculated (-11.6 to 5.8) was not significant but the limits of agreement were high with significant underestimation of %BF rendering the equation invalid. An explanation for these results could be the criterion method that was used which was isotope dilution. No results have been stated for validation against DEXA. In our study bias was not significant and more than 75% of the sample lied between a 5% variation as compared to DEXA. In 2010 a study was conducted in Hong Kong where children aged 9-19 were recruited and skin fold thickness were validated with subsequent formation of new equations[19]. However again the criterion used was air displacement plethysmography with only a subset validation with DEXA. They found significant difference between %BF estimated by the Slaughter equation although the absolute value was only of 1.52%. This was negated by our study which could be explained by the criterion method as being different and a significant difference in ethnicity. To the best of our understanding this is the first study conducted in Pakistani pediatric population to determine the correlation and validation of skinfold thickness equations for estimation of body fat composition using DEXA as gold standard.

LIMITATIONS:

Due to the addition of 2 schools the effect of institution was added as a result of which the formation and validation of fat mass equation for our population could not be assessed as the control group and validation group had significant differences between them.

As the age range was wide, different levels of maturity needed to be assessed by the Tanner staging.

Data for ethnic origin and socio-demographics was not held in consideration.

Furthermore the cut-off for accuracy to be deemed significant was 90% which was an arbitrary figure and not used previously. However the Slaughter equation revealed a figure of 98.4% which is very high.

In post hoc analysis regression analysis was performed for checking the line of identity in measured versus predicted fat mass and percent body fat. Although the Dezenberg and Goran equations revealed higher correlations with intercept not statistically different from 0 they were showing significant differences in fat mass prediction. While the Slaughter equation was showing an intercept different from 0 it was fulfilling all other criteria and we believe that if studies with larger sample size and accounting for ethnicity and maturity specific coefficients are performed it will stand its robustness.

CONCLUSIONS

We conclude that the Slaughter equation for estimating %BF showed reasonably strong validation with DEXA. However for interchangeable use with DEXA further studies with larger sample sizes and maturity considerations need to be conducted.

However, for estimation of fat mass, the previously formulated equations by Goran and Dezenberg revealed significant difference in our population. We do recommend further cross-validation studies for developing and validation of skin fold equation specific to Pakistani pediatric population.

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CONTRIBUTORSHIP STATEMENT:

All authors have substantially contributed to conception and design of study, acquisition of data, analysis and interpretation of data and preparatio of masnuscripty.

CONFLICT OF INTEREST:

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DATA SHARING STATEMENT

We are read to share data with the editorial board as per BMJ policy. No additional unpublished data is available.

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Table 1: Baseline Demographic and measured values of studied subjects

Total subjects: 99		
	Males = 54 mean (SD)	Females = 45 mean (SD)
Age (yrs)	13.8 (2.0)	14.13(2.2)
Weight (kg)*	41.7 (12.5)	43.6 (8.2)
BMI (kg/m^2)*	17.7 (3.5)	19.0 (3.4)
SC-SF (mm)*	9.9 (6.1)	14.1 (5.4)
T-SF (mm)*	11.4 (7.1)	16.0 (6.1)
FM by DZ-Eq	8.9 (5.4)	12.7 (4.1)
FM by G-Eq	20.7 (4.1)	28.6 (3.2)
%BF BY SL-Eq	10.7 (9.3)	13.3 (5.1)
DEXA FM*	8.3 (5.6)	10.2 (3.2)
DEXA %BF*	19.1 (7.6)	29.6 (5.1)

*: p value <0.05
BMI: Body Mass Index
SC-SF: Subscapular skinfold
T-SF: Tricep Skin Fold
FM: Fat Mass
%BF: Percentage Body Fat
DZ-Eq: Dezenberg Equation
G-Eq: Goran Equation
SL-Eq: Slaughter Equation

Table 2: Correlation coefficients calculated for prediction equations of DZ, G and SL for fat mass (FM) and percentage body fat (%BF). All equations showed good correlations with estimated fat mass and percentage body fat using DEXA showing significant p-values of <0.0001.

Equations	R^2	SEE
• DZ-FM	0.8	2.45
• G-FM	0.86	2.06
• SL-% BF	0.76	3.73

SEE: Standard Error of Estimate
DZ-FM: Dezenberg Fat Mass
G-FM: Goran Fat Mass
SL %BF: Slaughter Percentage Body Fat

Table 3: Validation criteria for prediction of % body fat and fat mass

	Slaughter	Goran	Dezenberg
Correlation	0.76	0.8	0.86
Accuracy	98.4	90.3	83.7
Precision	0.37 (p =0.4)	-1.26 (p<0.0001)	1.48 (p<0.0001)

According to all 3 criteria the Slaughter equation showed consistency good correlation, very high accuracy and good precision using paired t-test.

LEGENDS

Figure 1: (1a) Bland Altman plot shows uniform distribution between %BF calculated by DEXA and Slaughter. Although limits of agreement wide more than 75% of sample is between 5% variation; (1b) shows significant overestimation of fat mass between DEXA and Goran equation with almost the entire sample above the zero levels; (1c) shows significant underestimation of fat mass with more than 60% of the sample lying below the 0 level in the Dezenberg equation.

Figure 2: (2a,b) regression analysis with line of identity shows the intercept at an angle of 45 degrees not significantly different from zero in both the Dezenberg and Goran equations. Fig 2c reveals the regression analysis with line of identity between Slaughter %BF and estimated %BF DEXA with intercept significantly different from zero. Fig 2d reveals intercept not significantly different from zero between the SL equation for %BF and FM estimated by DEXA.

Correlation of skin folds thickness and validation of prediction equations using DEXA as gold standard for estimation of body fat composition in Pakistani children

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ABSTRACT

Objective: to determine the correlation between Dual Energy X-ray Absorptiometry (DEXA) and skinfold thickness equations (SKF) for estimation of body fat composition in secondary school children and validation of prediction equations by Slaughter, Goran and Dezenberg.

Design: Cross sectional analytical study.

Setting: Joint Commission for International Accredited (JCIA) tertiary care hospital of Karachi, Pakistan from January 2010 till May 2010.

Participants: The study was approved by Hospital's ethical review committee. Written and verbal consents were obtained from principals of two schools and parents of 99 children (mean age of 14 ± 1.89 years; min-max 9-19 years; 54 male and 45 female) accrued in study. DEXA scan was acquired and skin fold thickness was measured at angle of the scapula, iliac crest and mid arm for bicep and triceps skin folds using Holtain calipers. Correlations were established between estimated fat mass (FM) and percentage body fat (%BF) calculated by DEXA and those predicted by prediction equations. On obtaining significant correlation of >0.5, overall accuracy, **precision** and bias was calculated.

Results: There was an overall increased adiposity in females with fat mass of 3.57 kg and %BF 6.2% higher than male counterpart ($p < 0.05$). Slaughter equation predicted %BF accurately with overall high accuracy, minimal bias and with good **precision** with DEXA. Dezenberg and Goran equations had significant bias in prediction of fat mass which was statistically significant with low level of accuracy of the Goran equation.

Conclusion: We conclude that Slaughter equation for estimating %BF showed reasonable validation with DEXA. Nevertheless further studies with consideration for maturity and ethnicity are warranted for better results. However, for estimation of fat mass, previously formulated

equations by Goran and Dezenberg showed significant difference in our population. We do recommend further studies for developing and validation of skin fold equations specific to Pakistani pediatric population.

STRENGTH AND LIMITATIONS: This is the first well designed and conducted study from a JCIA accredited tertiary care Hospital (Aga Khan University Hospital), Karachi Pakistan. In this study various well known equations for estimation of percentage body fat and fat mass in Western population have been validated for Pakistani children. Relatively smaller sample size is the basic limitation of this research output which warrants and provokes local researchers to do further studies in future.

KEY WORDS: Pediatric Obesity; DEXA; percentage body fat; fat mass; Slaughter; Goran; Dezenberg; skin fold thickness equations

INTRODUCTION

Obesity is now being reported as the new world epidemic afflicting more than1 billion adults globally[1]. According to WHO reports a great number of children (22 million) are afflicted with obesity with a great proportion in the United States, however the developing nations are not to be spared[2]. Analysis of the national health survey in Pakistan for adults reveals that lower BMI thresholds were suggested than those recommended in western population based on their association with outcome of chronic disease[3]. Same situation exists in children. Retrospective analysis of data in Britain revealed that there was an increased risk of obesity in Afro Caribbean and Pakistani girls whereas Indian and Pakistani boys were found to be at higher risk for overweight[4]. In another study it was reported that application of international standards did not provide a reliable result due to significant difference in height at the same ages of children belonging to the Asian and African descent as compared to standard population who showed a greater proportion of central obesity[5]. Based on these results emerged the concept of measuring body fat composition as standardization of BMI as an obesity measurement index across different ethnicities remains to be formulated.

Body fat composition is measured in children using dual energy x-ray absorptiometry (DEXA) as gold standard. Multiple validations of this technique with chemical composition, total body weighting and hydro densitometry have been performed[6-9]. Due to its very small amount of radiation and short procedure time it is ideal for use in children.[10]

However it is not suitable for use in field work as it is not widely available and involves transportation of participants to tertiary care settings. Another method that has been validated

and widely used in the west is prediction equations based on anthropometric indices such as skinfold thickness of the biceps, triceps and abdominal wall as well as waist circumference and waist hip ratios. **The first equations that have been produced have initially derived by Slaughter et al [11]. In a sample of 310 children and adults age 8-29 yrs. prediction equations were cross validated against under body weighting using 4 compartment model, bone mineral studies and body water dilution. Special consideration for ethnicity and maturations was given. Multiple studies have tried to validate these equations in their population and on failing to do so derive their own equations. In a study by Goran et al they have previously cross validated DEXA against pig carcasses in the pediatric weight range and then after adjusting fat mass values with coefficient they have derived equations in prepubescent children by identifying the best predictors for fat mass and deploying them in a prediction equation[12].**

As a second part of the Goran study, Dezenberg et al also tried to validate the Slaughter and Goran equations in two different cohorts (the Alabama and the Vermont cohort) across multiple criteria and on failure to do so went ahead and formulated their own equations by identifying best predictors [13]. They too also came up with the conclusion that although much work has been done larger cohorts with different ethnicities is needed to establish the authenticity of prediction equations for widespread use in field work. As availability of DEXA is limited to larger centers it is imperative to validate skinfold thickness equations for measurement of obesity in general population.

The purpose of this study was to determine the correlation between DEXA and skinfold thickness equations for estimation of body fat composition in secondary school children and

validation of prediction equations (Slaughter, Goran and Dezenberg equations) against gold standard of DEXA by determining agreement and accuracy.

MATERIAL AND METHODS

This was a cross sectional analytical study conducted in Nuclear Medicine section, Department of Radiology, The Aga Khan University Hospital (AKUH), Karachi, Pakistan from Jan 2010 to May 2010-. The study was approved by AKUH ethical review committee.

The study population consisted of children enrolled form Class five till matriculate in the academic year from August of 2009 till May of 2010 form 2 community based schools belonging to the middle class strata. After the initial approval from the principal and obtaining school consent, a briefing lecture was delivered to the children with diagrammatic presentation of the machine and skin fold calipers. Thereafter assent forms to be signed by the children themselves and consent forms to be signed by the parents were distributed amongst the children. Furthermore phone numbers were obtained from the school administration and parents called to further explain the procedures and obtain verbal consent as well. All children whose parents gave consent were enrolled and those children from whom consent could not be obtained were excluded.

The sampling strategy was convenient sampling where consecutive children were enrolled till completion of sample size. A sample size of 44 subjects was needed to achieve 90% power to detect a change in slope from 0.0 under the null hypothesis to 0.45 under the alternative hypothesis when the standard deviation of the Triceps skin fold is 5.90, the standard deviation of Fat Mass by DEXA is 5.90, and the two-sided significance level is 0.05. These values were obtained from a study performed in children aged 4-10 years[14]. However since 2 schools were

included, by considering a design effect of 2, the required sample size was calculated as 88 subjects.

Data collection was performed on a designed questionnaire for the study. It included the socio-demographic data of the subject and all independent variables. Training of the principal investigator (PI) and the data collector was done according to the National Health & Nutrition Examination Survey (NHANES) manual[15]. A senior radiologist trained the data collectors for identification of the anatomical surface landmarks where measurements of the skin fold were to be obtained namely the angle of the scapula, the iliac crest and the midpoint of the arm for measuring bicep and triceps skin folds. Intra and inter observer variability was assessed for 10 sets of reading and readings with a discrepancy for more than 0.4 mm were retaken and the procedure revised. Abdominal and hip circumference was measured using a measuring tape according to the described technique and a difference of more than one centimeter was considered unacceptable.

DEXA scan was performed on Hologic Discovery-A QDR series, USA. The parameters of the scan were alternating fan beam radiations of 140 kV and 100 kV with a scan width of 26.4 inches and scan length of maximum 77 inches (195 cm). The participant was asked to change in a hospital gown and asked to lie straight on the table with hands by the side palms facing down away from the thighs and look at the ceiling to maintain head position. The body was positioned within the scan parameters marked on the table. The feet were rotated inwards so that the toes touched each other and taped so that the position was maintained. The method of performing the analysis of the estimation for %BF and total fat mass was done in the pediatric whole body scan mode for children up till 12 years of age. For children aged between 12 years onwards the adult

scan mode was used for the calculation of body fat composition parameters. Reliability between scans was achieved with phantom calibration.

The measure of fat mass and %BF calculated by DEXA were received as a printout from the scanner and values were entered in the study performa. Prediction of the same parameters via preformed prediction equations (Slaughter's, Goran and Dezenberg) using skinfold thickness and anthropometric measures were also made.

Body measurements:

All body measurements were taken in accordance with the protocol described for anthropometric measurements in the National Health & Nutrition Examination Survey-3[15]. Body measurements were taken on the right side of the body.

Anthropometric measurements were taken using specially designed calipers (Holtain Calipers). The measurements included weight, triceps skinfold thickness and sub scapular skinfold thickness.

Weight was measured using a Tanita TBF 305 body fat analyser and weighing scales (Tanita UK Ltd, Yewsley, Middlesex, UK).

Measuring skin fold thickness:

Skin fold measurements were obtained in various areas of the body such as the anterior and posterior aspect of the upper arm corresponding to the *Bicep* and *triceps* SF.

***Sub scapular SF* was obtained by measuring the subcutaneous tissue beneath the shoulder blade.**

***Suprailiac SF* was obtained by measuring the fold above the pelvic bone laterally.**

***Abdominal SF* was measured just adjacent to the umbilicus.**

***Abdominal circumference* was measured using a measuring tape at the level of the umbilicus.**

***Hip circumference* was measured at the maximum curvature of the hips.**

Skinfolds measurements were taken to the nearest 0.1 millimeter. All skinfold measurements were done in duplicate (i.e., twice by the same investigator) since these measures have the most variability. Skinfold thickness was measured by Holtain calipers. The procedure was thoroughly explained to the children and the use of the caliper was demonstrated on the child's palm.

The following prediction equations were used:

***Fat mass* is obtained as amount of fat calculated in kilograms predicted by the DZ and G equations**

Dezenberg equation:

FM (kilograms) = 0.342 × body mass (kg) + 0.256 × triceps SF (mm) + 0.837 × sex (1= boy, 2 = girl) - 7.388

Goran equation:

FM (kg) = 0.23 (sub scapular SF) + 0.18 (weight) + 0.13 (triceps SF) – 3.0

***Percentage body fat* is obtained as a percentage of body fat predicted by the Slaughter equation.**

For boys:

%BF = 1.21 [sum of triceps and subscapular skinfolds in mm] - 0.008 [9sum of triceps and subscapular skinfolds in mm) ^{2}] - 1.7 (coefficient for white Prepubescent boys in boys with sum of triceps and subscapular skinfolds < 35 mm);

If sum of triceps and subscapular skinfolds > 35 mm,

%BF = 0.783[Sum of triceps and subscapular skinfolds mm] - 1.7 in boys;

For girls:

%BF = 1.33 [sum of triceps and subscapular skinfolds _mm] - 0.013 [(sum of triceps and subscapular skinfolds in mm) ^{2}] + 3.0 (coefficient for white girls) in girls with sum of triceps and subscapular skinfolds < 35 mm;

Or if sum of triceps and subscapular skinfolds > 35 mm,

%BF = 0.546 [sum of triceps and subscapular skinfolds in mm] + 9.7 in girls

Data for all the independent variables was collected by the PI and assisting technician in the Dexa room. Two readings were taken for height, weight, abdominal and hip circumference, triceps, and subscapular and suprailiac skin fold thicknesses **and the average measurement for both the readings was taken as the final reading**. If a difference of more than 0.3 mm was recorded the readings were retaken. Calibration of the calipers was performed periodically according to the calibration block provided by the manufacturers. The stadiometer and weighing scale were also calibrated and checked for correct position of zero before each data set recording.

Statistical analysis: Descriptive statistics were calculated for the data with mean and standard deviations for all continuous variables. Proportions were described for categorical data such as gender and independent T-test was applied to assess significant difference among gender.

The next step of the analysis was to validate the equation for fat mass and percentage body fat described by Goran, Dezenberg and Slaughter. In order to do this initially correlations were established between estimated FM and %BF calculated by DEXA and that predicted by using the 3 equations. On obtaining significant correlation of > 0.05, precision and bias was calculated.

Accuracy was defined as the degree of closeness of predicted fat mass and percent body fat obtained from the equations to their actual criterion value estimated by DEXA and would be considered significant if more than 90%. This was done by obtaining the overall mean of fat mass and %BF by DEXA as well as the Dezenberg, Goran and Slaughter equations **separately**. **Individual accuracies were then calculated by subtracting the predicted mean of percent body fat and fat mass obtained from the equations from the mean value of fat mass and percentage body fat calculated by DEXA.** The difference was then expressed as a percentage of the mean of the **estimated DEXA value for the 3 equations.**.

Precision was defined as the variation between the predicted and estimated values of %BF and FM within the data and was assessed as the average of the difference between FM and %BF measured by DEXA and that predicted by the equations and plotting the mean of errors against 0 using **paired samples** t-test.

Bland Altman plots were also generated for evaluation of **agreement and bias**. Based on the above mentioned criteria the equation for %BF by Slaughter and for fat mass by Dezenberg and

Slaughter were rendered valid or rejected. **Post hoc regression analysis was also performed with the line of identity against predicted versus estimated fat mass and percent body fat.** A commercial Statistical Package for Social Sciences (SPSS Version 17) was used for these measurements.

RESULTS

Descriptive analysis and gender difference:

Ninety nine (n= 99) subjects were registered with a mean age of 14 ± 1.89 years (min-max 9-19 yrs) with 54 male and 45 female subjects. There was an evidence of overall increase in the adiposity of females as compared to males. All skin fold measures as well as fat composition variables were significantly higher in girls with p-value < 0.05. After assigning the binary variables it was observed that there was a difference of 3.57 kg fat mass present in excess in females as compared to the males as calculated by DEXA (p-value <0.0001). Significant differences were also obtained for %BF which was 6.2 % more in females as compared to males (p-value < 0.05). (Table 1)

After obtaining baseline data, the equations were applied to the sample for prediction of fat mass by Dezenberg and Goran as well as %BF by Slaughter. In the linear regression analysis strong correlations were obtained between the predicted values and the criterion values by DEXA with p-value<0.0001. (Table 2).

Validation of equations:

Overall accuracy:

The overall accuracy for the Slaughter equation was 98.4% which was very high and Dezenberg equation had an overall accuracy of 90.3% which was just within the cut-off designed. Goran equation showed weak overall accuracy of 83% which did not fall within the cutoff defined.

Variation/Precision of equations

No significant difference was obtained between the Slaughter equation predicting %BF and that estimated by DEXA with a difference of 0.37 kg (p-value **0.425**). However, significant differences for predicting fat mass were obtained between DEXA and Dezenberg. The equation underestimated the fat mass with a bias of 1.26 kg which was statistically significant (p-value <0.0001). While Goran equation overestimated the fat mass with a bias of 1.48 kg which was statistically significant with a p-value of <0.0001

Calculating Bias:

Limits of agreement were evaluated using Bland Altman analysis. The slaughter equation revealed that more than 75% of the data was within the first standard deviation.

Thus in view of the above criteria the Slaughter equation fulfilled all the criteria for predicting %BF accurately as the overall accuracy is high with minimal variation which is not statistically

significant and showing good **precision** with %BF measured by DEXA. Furthermore more than 75% of the readings were within 5% of the estimated DEXA values.

The Dezenberg and Goran equations had significant variation in prediction of fat mass which was statistically significant with low level of accuracy of the Goran equation. A summary of the results is shown in Table 3

Post hoc regression analysis revealed that both the Goran and Dezenberg equation did not reveal an intercept significantly different from zero. The Slaughter equation did not reveal an intercept significantly different from zero against estimated fat mass however the intercept was significantly different from zero when it was assessed against estimated percent body fat (Fig 1).

DISCUSSION

The onset of obesity in the pediatric population has shown early precedence of atherosclerotic changes and diabetes mellitus with the onset of metabolic syndrome thus rendering obesity as a major health concern. Thus with this background we set out to conduct a study for the measurement of obesity in our pediatric population. A new technique of predicting body fat composition is by prediction equations (Salughter[11], Goran[12], and Dezenberg[13] equations) which have been developed for the western world. We attempted to validate these equations to predict body fat composition in our pediatric population which comprised of fat mass and %BF using DEXA as the gold standard. In this study only the Slaughter equation was validated for predicting %BF with a good correlation and high accuracy (98.4%). No significant bias was seen amongst the predicted and estimated method for measuring %BF. These equations have been validated in different studies and used widely. In a study carried out by Janz in 122 subjects of pre and post pubescent children ranging from 8-17 years very high intra class correlations (0.98-0.99) and high validity correlations of (0.79-0.99) were achieved[16]. In another study conducted in Zaragoza, Spain carried out on 237 adolescents it was found that the Slaughter equations had the narrowest limits of agreement as compared to the other equations and did not show statistically significant bias[17]. Our results have been similar with the equation showing validity across all criteria.

In the study by Goran et al, after failing to validate the Slaughter equation they derived an equation for their own sample. It was reported that all skin folds showed higher correlations with fat mass rather than percent body fat so equation for the prediction of fat mass was generated. Another method of calculating fat composition was also deployed in this study using bioelectrical impedance analysis. They found the variables of triceps, sub scapular and weight significantly correlating. The equation was further cross validated on 1/3rd of the sample according to the criteria of Lohman which consisted of assessing the difference between the predicted and estimated fat mass as well as minimal increase of the SEE. Only the R^2 was reported of 0.88.

This equation was then further cross validated in the study by Dezenberg. In his study after initially failing to validate the Goran and Slaughter equation, a prediction equation was formulated across a sample of different ethnic cohorts consisting of both black and white population. The predictors considered significant were weight; triceps skin fold, gender, ethnicity and abdominal skin fold. However since we were not evaluating ethnicity, the equation adopted in this study consisted only of weight, triceps skin fold and gender predictors. A significant point of concern in this study was the fact that regression analysis was performed between percentage body fat as predicted by Slaughter against fat mass as predicted by dexa. Since both variables were in different units the inability to validate the equations appeared inevitable. In our study adopting the approach of Dezenberg et al we achieved good correlation with the intercept not significantly different from zero with Slaughter equations predicting percent body fat and fat mass in kilograms estimated by dexa (fig 2d) however the regression analysis between predicted percent body fat and that

estimated with dexa shows an intercept significantly different from zero. Despite this as multiple validation criteria were applied and the Slaughter equation maintained robustness across all of them it can be used with some considerations of maturity specificity and ethnicity in our population.

Our results were similar to the results by Eisenmann who evaluated a number of equations along with bioelectrical impedance analysis. In his study he found that the Slaughter equations consisting of triceps and subscapular skinfold were showing better limits of agreement and reduced bias there only one of the slaughter equations was adopted. Significant differences were assessed between the methods for evaluating body fat composition by DEXA and SF equations, bioelectrical impedance.

A few issues are worthy of mention here. These results could be explained by the application of coefficients that were derived for the western population. Therefore determining coefficients for the predictors used in the Dezenberg equation and deriving a prediction equation that is sample specific could possibly yield better results.

Another reason for inability to validate the DZ equation might be that our sample consisted of both pre and post pubescent children. The application of maturity specific coefficients by performing the Tanner staging might have different results as these were conducted on children who were prepubescent. Furthermore data regarding the ethnic origin and socio demographic status was also not applied. However all children were living in the same town going to community center schools with a middle class status.

The Goran equation also was established on a cohort of children whose mean age was much lower than that of our sample. Furthermore gender was not accounted for in the equation we used whereas in our population significant differences were found between boys and girls for fat mass as well as all skin folds.

Very few studies have been done regionally in the South Asian population. In a study conducted in Sri Lanka in 282 children ages ranging from 5-15 years were tested for predicted %BF using multiple equations as well as Slaughter equation[18]. Although the bias calculated (-11.6 to 5.8) was not significant but the limits of agreement were high with significant underestimation of %BF rendering the equation invalid. An explanation for these results could be the criterion method that was used which was **isotope dilution**. No results have been stated for validation against DEXA. In our study bias was not significant and more than 75% of the sample lied between a 5% variation as compared to DEXA. In 2010 a study was conducted in Hong Kong where children aged 9-19 were recruited and skin fold thickness were validated with subsequent formation of new equations[19]. However again the criterion used was air displacement plethysmography with only a subset validation with DEXA. They found significant difference between %BF estimated by the Slaughter equation although the absolute value was only of 1.52%. This was negated by our study which could be explained by the criterion method as being different and a significant difference in ethnicity. To the best of our understanding this is the first study conducted in Pakistani pediatric population to determine the correlation and validation of skinfold thickness equations for estimation of body fat composition using DEXA as gold standard.

LIMITATIONS:

Due to the addition of 2 schools the effect of institution was added as a result of which the formation and validation of fat mass equation for our population could not be assessed as the control group and validation group had significant differences between them.

As the age range was wide, different levels of maturity needed to be assessed by the Tanner staging.

Data for ethnic origin and socio-demographics was not held in consideration.

Furthermore the cut-off for accuracy to be deemed significant was 90% which was an arbitrary figure and not used previously. However the Slaughter equation revealed a figure of 98.4% which is very high.

In post hoc analysis regression analysis was performed for checking the line of identity in measured versus predicted fat mass and percent body fat. Although the Dezenberg and Goran equations revealed higher correlations with intercept not statistically different from 0 they were showing significant differences in fat mass prediction. While the Slaughter equation was showing an intercept different from 0 it was fulfilling all other criteria and we believe that if studies with larger sample size and accounting for ethnicity and maturity specific coefficients are performed it will stand its robustness.

CONCLUSIONS

We conclude that the Slaughter equation for estimating %BF showed reasonably strong validation with DEXA. However for interchangeable use with DEXA further studies with larger sample sizes and maturity considerations need to be conducted.

However, for estimation of fat mass, the previously formulated equations by Goran and Dezenberg revealed significant difference in our population. We do recommend further cross-validation studies for developing and validation of skin fold equation specific to Pakistani pediatric population.

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CONFLICT OF INTEREST:

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Table 1: Baseline Demographic and measured values of studied subjects

Total subjects: 99	Males = 54 mean (SD)	Females = 45 mean (SD)
Age (yrs)	13.8 (2.0)	14.13(2.2)
Weight (kg)*	41.7 (12.5)	43.6 (8.2)
BMI (kg/m^2)*	17.7 (3.5)	19.0 (3.4)
SC-SF (mm)*	9.9 (6.1)	14.1 (5.4)
T-SF (mm)*	11.4 (7.1)	16.0 (6.1)
FM by DZ-Eq	8.9 (5.4)	12.7 (4.1)
FM by G-Eq	20.7 (4.1)	28.6 (3.2)
%BF BY SL-Eq	10.7 (9.3)	13.3 (5.1)
DEXA FM*	8.3 (5.6)	10.2 (3.2)
DEXA %BF*	19.1 (7.6)	29.6 (5.1)

*: p value <0.05
BMI: Body Mass Index
SC-SF: Subscapular skinfold
T-SF: Tricep Skin Fold
FM: Fat Mass
%BF: Percentage Body Fat
DZ-Eq: Dezenberg Equation
G-Eq: Goran Equation
SL-Eq: Slaughter Equation

Table 2: Correlation coefficients calculated for prediction equations of DZ, G and SL for fat mass (FM) and percentage body fat (%BF). All equations showed good correlations with estimated fat mass and percentage body fat using DEXA showing significant p-values of <0.0001.

Equations	R^2	SEE
• DZ-FM	0.8	2.45
• G-FM	0.86	2.06
• SL-% BF	0.76	3.73

SEE: Standard Error of Estimate
DZ-FM: Dezenberg Fat Mass
G-FM: Goran Fat Mass
SL %BF: Slaughter Percentage Body Fat

Table 3: Validation criteria for prediction of % body fat and fat mass

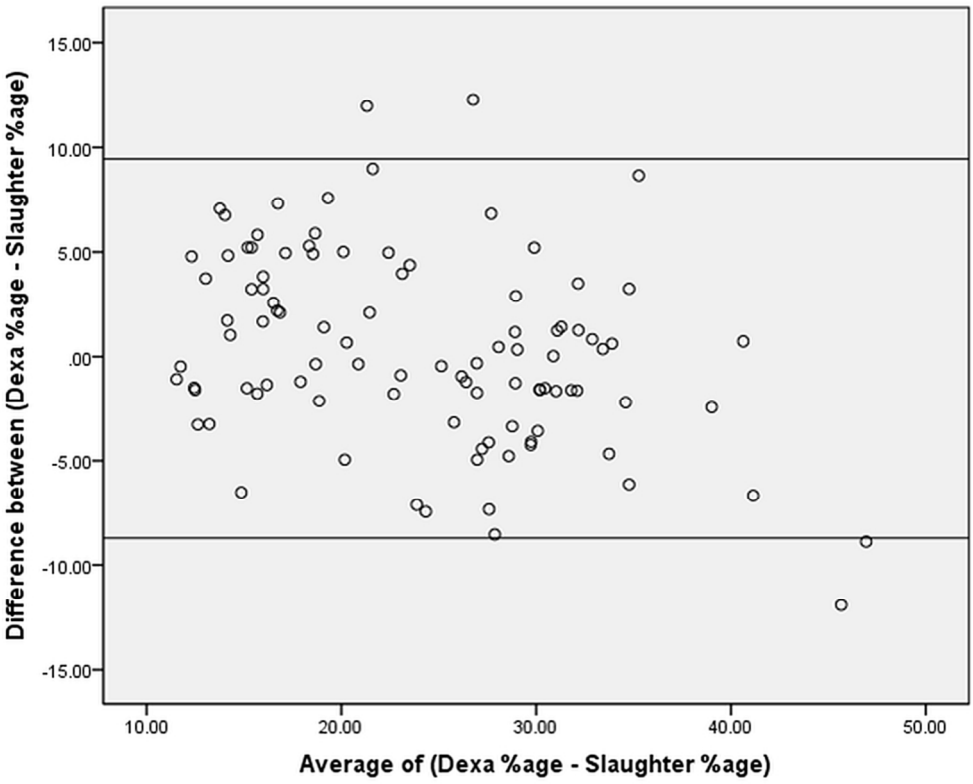
	Slaughter	Goran	Dezenberg
Correlation	0.76	0.8	0.86
Accuracy	98.4	90.3	83.7
Precision	0.37 (p =0.4)	-1.26 (p<0.0001)	1.48 (p<0.0001)

According to all 3 criteria the Slaughter equation showed consistency good correlation, very high accuracy and good precision using paired t-test.

LEGENDS

Figure 1: (1a) Bland Altman plot shows uniform distribution between %BF calculated by DEXA and Slaughter. Although limits of agreement wide more than 75% of sample is between 5% variation; (1b) shows significant overestimation of fat mass between DEXA and Goran equation with almost the entire sample above the zero levels; (1c) shows significant underestimation of fat mass with more than 60% of the sample lying below the 0 level in the Dezenberg equation.

Figure 2: (2a,b) regression analysis with line of identity shows the intercept at an angle of 45 degrees not significantly different from zero in both the Dezenberg and Goran equations. Fig 2c reveals the regression analysis with line of identity between Slaughter %BF and estimated %BF DEXA with intercept significantly different from zero. Fig 2d reveals intercept not significantly different from zero between the SL equation for %BF and FM estimated by DEXA.



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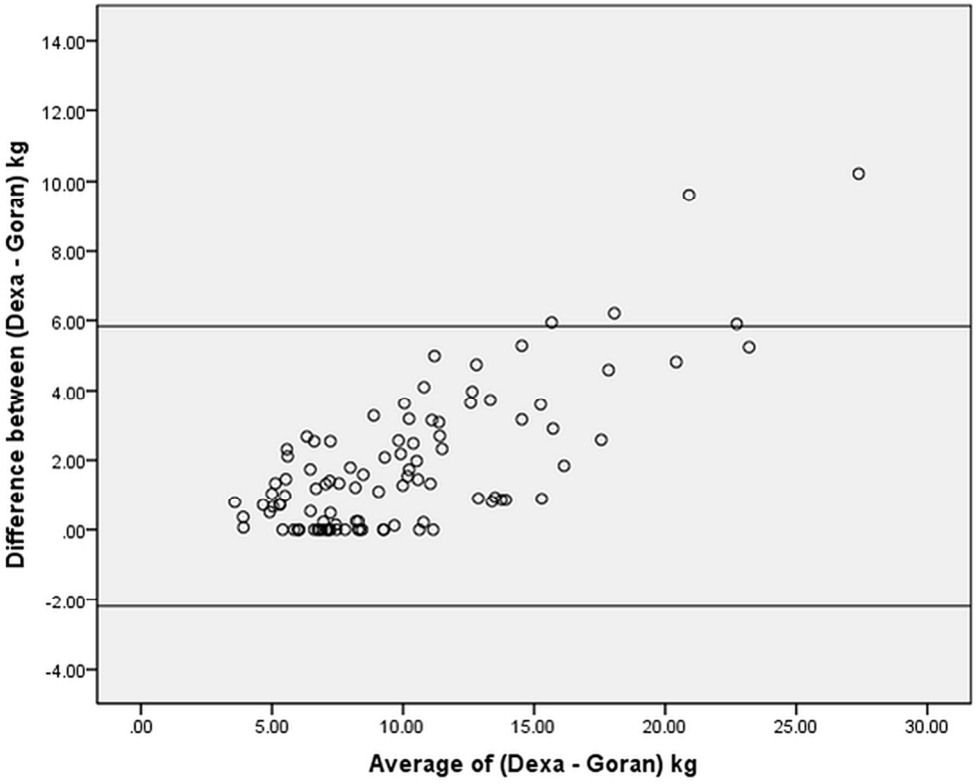

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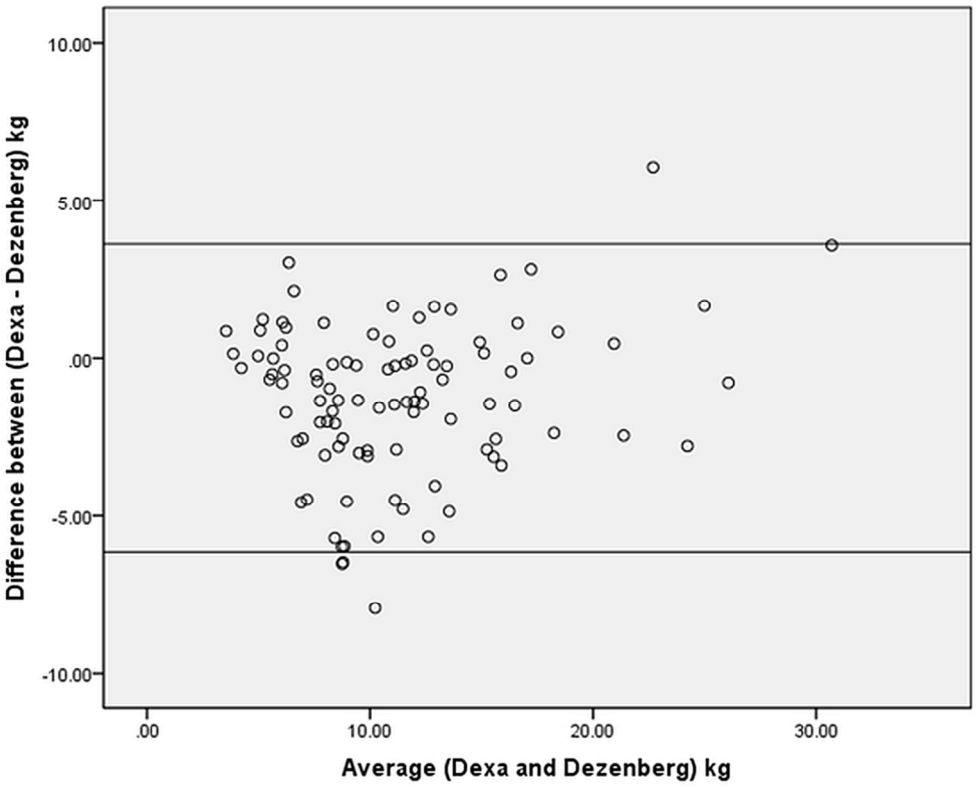

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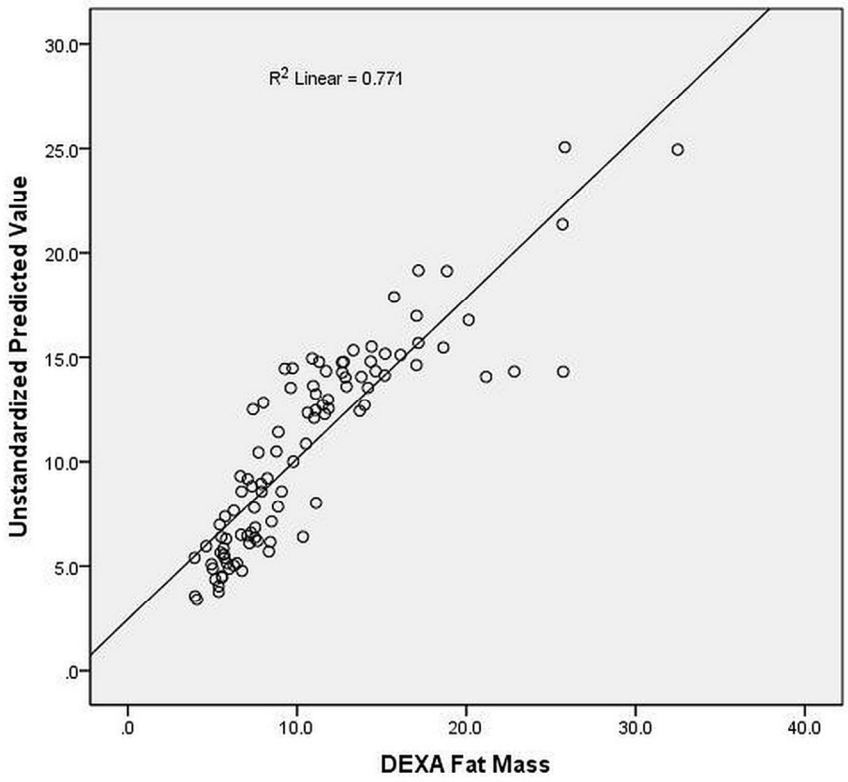

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