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Predicting visceral adipose tissue by MRI using DXA and anthropometry in adolescents and young adults

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Abstract

Objective—Accumulation of intra-abdominal (visceral) adipose tissue, independent of total adiposity, is associated with development of metabolic abnormalities such as insulin resistance and type-2 diabetes in children and adults. The objective of this study was to develop prediction equations for estimating visceral adiposity (VAT) measured by magnetic resonance imaging (MRI) using anthropometric variables and measures of abdominal fat mass from DXA in adolescents and young adults.

Methods—Cross-sectional data was collected from a multiethnic population of seventy males and females, aged $12-25$ years, with BMI ranging from $14.5-38.1$ kg/m². Android (AFM; android region as defined by manufacturers instruction) and lumbar L1-L4 regional fat masses were assessed using DXA (GE Lunar Prodigy; GE Lunar Corp, Madison, WI, USA). Criterion measures of intra-abdominal visceral fat were obtained using single-slice MRI (General Electric Signa Model 5x 1.5T) and VAT area was analyzed at the level OF L4–L5. Image analysis was carried out using ZedView 3.1.

Results—DXA measures of AFM $(r=0.76)$ and L1-L4 $(r=0.71)$ were significantly $(P<0.0001)$ correlated with MRI-measured VAT. DXA AFM, together with gender and weight, explained 62% of the variance in VAT (SEE=10.06 cm²). DXA L1-L4 fat mass with gender explained 54% of the variance in VAT (SEE=11.08 cm²). Addition of the significant interaction, gender \times DXA fat mass, improved prediction of VAT from AFM (R_{adj}^2 =0.61, SEE=10.10cm²) and L1-L4 $(R_{\text{adj}}^2=0.59, \text{SEE}=10.39 \text{cm}^2).$

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Author contributions: The authors responsibilities were as follows- TGL and SBG: designed the study and were the Project Investigators; VRL and RMB: were project coordinators, and collected data; RMB collected all DXA scans; TS: collected and analyzed MRI-VAT data; DRL: analyzed all DXA and MRI analysis under the supervision of RMB and VRL; DRL: conducted statistical analysis, interpreted results of experiments, drafted the manuscript and prepared tables; SBG edited and approved final version of the manuscript; and all authors: contributed to revisions of the manuscript.

Conclusion—These results demonstrate that VAT is accurately estimated from regional fat masses measured by DXA in adolescents and young adults.

Keywords

Android fat mass; Visceral Adiposity; body composition; dual-energy x-ray absorptiometry; MRI

Introduction

Excess intra-abdominal (visceral) adipose tissue, independent of total adiposity, is a major determinant of the metabolic syndrome $[1-3]$, insulin resistance $[2, 4, 5]$, cardiovascular disease (CVD) [2, 6, 7], and type-2 diabetes [2] in children, adolescents [8, 9] and adults. At present, reliable imaging techniques for measuring visceral abdominal adiposity include magnetic resonance imaging (MRI) and computed tomography (CT), which directly measure intra-abdominal adipose tissue (IAAT), allowing for quantification of several fat depots with a degree of accuracy comparable to chemical analysis [1, 10–13]. However, both are expensive, and access is often limited. Also, the radiation exposure from CT is high, limiting its use in children and longitudinal designs [14]. Lower cost, accessible methods for safely estimating IAAT, especially in children and youth, are needed.

Indirect methods, including the use of a variety of anthropometric measures, eg waist circumference (WC), hip circumference (HC), waist-to-hip ratios (WHR) and skinfolds [1, 11, 15–17], have also been used to estimate visceral adiposity (VAT). These methods are the most common because they are practical, portable, noninvasive, and inexpensive, and they can be used to monitor changes in diverse clinical settings. Nevertheless, the accuracy of anthropometric variables is limited as variations in body build and the percentage and distribution of adipose tissue, which varies with age, sex, and ethnicity [18], confounds the relationship with VAT [11, 19]. While WC and WHR have been used as convenient surrogates for central adiposity [4, 11, 16, 20, 21] and are useful for characterizing fat distribution, they do not accurately detect small changes in VAT that can occur over time. Moreover, because a standard measurement site has not been adopted, measurements of WC or WHR are often not comparable [10].

Dual-energy X-ray absorptiometry (DXA), which has emerged as a criterion method for assessing regional and whole body soft tissue composition [10, 11, 14, 22], is less invasive, less expensive and more accessible than CT, and involves only minimal exposure to ionizing radiation [1, 10]. Previous studies have demonstrated that DXA-derived trunk fat mass [11] and abdominal tissue mass in the L1–L4 area are associated with abdominal fat mass and VAT in adults [1, 23]. When compared to WC, DXA can predict fat mass with greater accuracy and reproducibility and may potentially serve as a useful tool for tracking small changes in abdominal fat during weight loss and maintenance therapies [1, 10]. Past studies to predict CT-measured VAT using DXA measures of trunk [11] or regional abdominal fat [1, 24, 25], have focused primarily on adults and its utility for estimating VAT in adolescents has not been established. Thus, the primary aim of this study was to develop an algorithm for accurately estimating VAT from DXA using MRI to obtain criterion measures of VAT. Prediction of VAT from a manually drawn region of interest (ROI) spanning the

abdomen (L1-L4) was compared to manufacturer's default regions (trunk and android regions) to assess whether prediction for the default ROI was as good as a manual ROI. Anthropometric variables were included in the analysis to determine whether prediction improved when anthropometry and DXA were combined.

Subjects and methods

Anthropometric characteristics and body composition assessments were completed on 70 males and females, 12–25 years of age, following the procedures described below. The protocol was approved by the University of Arizona Human Subjects Protection Committee, and the study was conducted in accordance with the Helsinki Declaration. Written informed consent was received from all subjects and the guardians of participants under the age of 18 years. Volunteers were excluded if they had a history of chronic disease (eg HIV/AIDS, congestive heart failure, unstable angina), or cancer; any implanted electronic medical equipment or external life support equipment; metal implants; jewelry that could not be removed; had taken medications that may affect body composition, fat distribution, or physical activity (ie growth hormone); had been diagnosed with a disease or condition that may affect body composition (ie Cushing's Syndrome, Type 1 or Type 2 diabetes, thyroid disorder) [17] or had learning disabilities that made it difficult to complete questionnaires, were unable to comply with assessment protocols, or unable to read and understand English. Females who were pregnant or nursing were also excluded. Individuals were also excluded if they had a fear of small-enclosed spaces or were unable to remain in a lying or sitting position for an extended period ($\overline{30}$ minutes) of time, as required by the MRI procedures.

At the initial visit, demographic data (i.e., age, gender, race/ethnicity) were obtained through questionnaires; anthropometric measurements, including weight, height, waist, and hip circumferences were also taken. Subjects were then scheduled for a whole body DXA scan and total body MRI scan. All measurements were completed within a 7-day period.

Anthropometry

Measures of body weight, standing height, and waist and hip circumference were obtained by a trained anthropometrist. Body weight was measured to the nearest 0.1 kg, using a calibrated digital weighing scale (Seca Model 770 scale, Hamburg, Germany), with subjects minimally clothed in light-weight swimwear or underwear. Standing height (stature) was measured to the nearest 0.1 cm with the shoes removed and the head in the Frankfort plane using a standard stadiometer (Shorr Height Measuring Board, Olney, MD). Waist circumference was measured to the nearest 0.1 cm with anthropometric tape placed at the midpoint between the lower rib margin and the iliac crest, and hip circumference was measured to the nearest 0.1 cm at the widest point over the greater trochanters. Each anthropometric variable was measured three times and the respective averages were used in the analysis.

Dual energy x-ray absorptiometry (DXA)

Soft tissue mass and composition, including total-body mass, total-body fat mass, and region specific abdominal fat mass, were assessed using dual-energy X-ray absorptiometry (DXA)

using the GE Lunar Prodigy (software Version 5.60.003) densitometer (GE Lunar Corp, Madison, WI, USA). Subjects were positioned following the standard manufacturer protocols. Participants were asked to lay supine with their arms resting by their sides (not touching the body), wrists pronated, and hands flat [22]. Subjects were scanned in light clothing or hospital gowns, with all artifacts removed from the scan area [10, 26]. All participants were scanned on the same machine, and DXA scan acquisition and review were performed by one of two certified technicians. The Lunar Prodigy was calibrated daily according to the standard procedures for maintenance and use as recommended by the manufacturer. DXA CVs for precision of whole body and regional soft tissue composition in our laboratory are <1 to 3% (22), similar to estimates reported from other laboratories [27, 28].

DXA regional analysis—Abdominal area regions of interest (ROIs) included android and L1-L4 regions. Android fat mass (AFM), available from the manufacturer's automated ROIs, is defined as the area enclosed between a demarcation above the iliac crest to the 20% mark of the total distance between the iliac crest and the base of the skull. The manually drawn L1-L4 ROI was chosen based on previous studies in adults [1, 26] and adolescents [22]. L1-L4 was defined via delineation of the lumbar spine region including the area bounded by the upper most border of the L1 to the lower most border of L4.

Magnetic resonance imaging

Abdominal VAT was estimated using whole-body magnetic resonance imaging (MRI). MRI was performed by an experienced technician using the General Electric Signa Model $5\times$ 1.5T MRI scanner. Subjects lay supine on the scanner bed, with their arms extended above their heads. Images in abdominal and thoracic regions were obtained with the subjects holding their breath. The scanning process was divided into two parts with the ischial tuberosity as the point of origin to divide the body into upper and lower sections. The lower body was scanned first, followed by the upper body. Total test time was approximately one hour. The total number of axial images taken across the abdominal area was determined relative to the participant's height (height/50mm; spacing between slice=50mm; field of view 480 mm (1.875 mm * 256 pixel); slice thick-ness=10.0 mm thickness). The single slice method was used to estimate the intra-abdominal visceral fat area $\text{(cm}^2\text{)}$. Images were analyzed using ZedView 3.1 (LEXI Corporation, Ltd., Tokyo, Japan ([http://www.lexi.co.jp/](http://www.lexi.co.jp/e_zedview.html) [e_zedview.html\)](http://www.lexi.co.jp/e_zedview.html). Protocol details have been published elsewhere [29]. Briefly, the software employed knowledge-based image processing to label pixels as fat and nonfat components using on the basis of the gray-level histograms of the images. Each slice was manually reviewed and VF area was analyzed at the level L4–L5. Voxels arising from fatty bowel content were deleted. VF in cm^2 was divided by 10 and rounded to derive VF_{level} [29]. The MRI scanner was calibrated daily according to the manufacturer instructions for maintenance and use.

Statistical analysis

Scatter plots were examined for outliers and skew-ness and kurtosis were calculated for all variables. Descriptive statistics were calculated for the entire sample. Bivariate relationships were estimated using Pearson's correlation coefficients (r) for continuous variables and

Spearman's rank order correlation coefficients (rho) for categorical variables. Fischer's Ztransformation test (FZT) was used to test correlation coefficients for differences between males and females. Stepwise multiple regression analyses were run to derive prediction equations for estimating MRI total VAT mass from DXA derived android or L1-L4 abdominal fat in combination with anthropometric measures. Other independent variables *considered* in the models included body weight, height, body mass index (BMI), waist circumference, age, gender (male = -1 , female = 1), and ethnicity (Asian=1, African American=2, Hispanic=3, White=4), and the interaction of gender with android and L1-L4 fat. The level of significance was set at *P*<0.05 (two-tailed). All analyses were performed using the Statistical Package for the Social Sciences for Windows, Version 18.0 (SPSS, Chicago, IL, USA).

Results

Descriptive characteristics for the entire sample and by sex are shown in Table 1. The sample was comprised of 35 males and 35 females (n=70) and included 2 Asians, 7 African Americans, 15 Hispanic and 46 non- Hispanic white subjects. The sample was a mixture of underweight (n=10), normal weight (n=43), overweight (n=15) and obese (n=2) individuals based on BMI. The mean weight for the entire sample was 64.4kg. Bivariate correlations between potential model covariates and fat masses are shown in Table 2. The coefficients did not differ (>0.05) between males and females, thus only results for the total sample are reported (table 2). Age (r=0.25; *P* <0.04), waist (r=0.58; *P* <0.0001), BMI (r=0.56; *P* ≤ 0.0001), and weight (r=0.42; *P* ≤ 0.0001) were significantly correlated with MRI estimates of VAT. Additionally, DXA measures of android fat mass (AFM) (r=0.76, *P* <0.0001) and L1-L4 (r=0.71, *P* <0.0001) were positively correlated to MRI-measured VAT; the relationship was slightly stronger with android fat mass than with L1-L4 ROI.

Development of prediction equations by subsection

Results for models predicting MRI VAT from DXA measures of fat mass are reported in Tables 3 and 4. Because AFM and L1-L4 are highly inter-correlated (r=0.982), they were tested in separate models for estimating VAT. DXA measures of anthropometric covariates (weight, height, BMI, waist) and demographic covariates (eg age, race, and gender) were then added (stepwise) to test their additional contributions (Tables 3 and 4).

The model using only DXA-derived AFM (*P* <0.0001) explained 57% of the variance in VAT by MRI. Results from stepwise regression showed that only gender $(P \le 0.013)$ was a significant predictor of VAT and inclusion of gender in the AFM model increased the R_{adi}^2 from 0.57 to 0.60 and reduced the SEE (cm²) from 10.70 to 10.30 cm². Addition of the anthropometric covariate, weight, $(P \le 0.046)$ resulted in a further improvement in the prediction of VAT, increasing the $R_{adj}^2=0.62$ (SEE=10.06 cm²) (Table 3). Further analysis suggested an interaction between gender and AFM (*P* <0.062). The final model to predict VAT using AFM, gender and the interaction term explained 61% of the variance in VAT $(SEE=10.10 \text{ cm}^2)$ (Table 3).

The model using only DXA-derived L1-L4 ROI ($P < 0.0001$) explained 50% of the variance in VAT. Inclusion of gender $(P<0.014)$ to the L1-L4 model increased the variance

explained by the model, increasing the $R_{adj}^2 = 0.54$, and reducing the SEE from 11.51 cm² to 11.08 cm² (Table 4); however, in L1-L4 models, no other anthropometric or demographic variables were significant predictors of VAT. Further analysis revealed a significant interaction between gender and L1-L4 (gender*L1-L4). The final model to predict VAT using DXA L1-L4, gender and the interaction term explained 59% of the variance, and reduced SEE to 10.39 cm².

Models including AFM were, overall, better at predicting VAT than L1-L4 ROI, and for any given model, AFM explained 2% to 7% more variance in VAT compared to L1-L4 ROI. The prediction of VAT was further improved when the interaction between gender and AFM or L1-L4 was included in the model.

Discussion

DXA-derived L1-L4 and android fat mass (AFM) were evaluated separately and with anthropometric variables to predict VAT. AFM and L1-L4 ROI had similar, significant correlations with VAT ($R=0.76$; $R=0.71$, $P < 0.0001$), and both provided more accurate prediction of VAT than anthropometry alone and anthropometry combined with demographic covariates (data not shown). Thus, DXA measures of AFM and L1-L4 have a clear advantage over anthropometry alone for predicting VAT. Demographic variables were also evaluated in models with AFM and L1-L4 fat mass as previous studies have shown that age, race and gender differences in fat distribution [30–32] might affect the relationship between AFM and L1-L4 VAT. In this sample, only gender was a significant predictor of VAT in models with AFM and L1-L4, and addition of body weight to models including gender and AFM (but not L1-L4 fat mass) reduced error and increased the variance explained in VAT. Inclusion of the interaction of gender with AFM or L1-L4 further improved the prediction of VAT.

The findings of this study agree with earlier studies in both adults and children which have investigated the combination of anthropometric measures and DXA estimates of regional adipose tissue distribution as potential correlates of VAT and intra-abdominal adiposity. While some studies have focused on standardized regions included in DXA software, others have delineated custom regions of interest and examined their relationship with VAT [14, 22].

Conventional assessment of trunk fat by DXA, for example, has been used to predict intraabdominal adiposity. In adult women [33] and pre-pubescent children [17], DXA derived trunk fat combined with anthropometric variables explained 81% ($SEE = 24.6$ cm²) [33] to 85% (SEE=8.9 cm²) [17] of the variance, respectively, in CT measures of IAAT. However, the accuracy of these DXA prediction equations is limited, because trunk includes the entire thoracic and abdominal areas rather than an anatomical region more closely aligned with VAT [17]. Hill and colleagues (2006) found that in their sample of overweight or obese women, DXA fat mass from manually drawn abdominal ROIs at 5 cm (r=0.70) and 10 cm (r=0.78) regions (above iliac crest), which were closer in proximity to IAAT measures from CT, were moderately correlated with IAAT (27). Inclusion of abdominal skinfolds with the DXA 10 cm ROI improved the amount of variance in IAAT (R=0.82), that could be

explained. Interestingly, Bertin and colleagues (2000) found that DXA abdominal fat estimates using a specially designed version of the software that accounted for intraabdominal cavity thickness (e.g. transverse internal diameter and transverse external diameter), combined with abdominal sagittal diameter, age, and waist circumference, resulted in strong correlations with CT-measured IAAT in obese men (r=0.88) and obese women (r=0.94) with an estimated error for the combined sample of men and women of 38.2 cm^2 [34]. Other studies in adults, examining ROIs at the L2 – L4 area combined with waist circumference, have shown similar correlations with IAAT ($r = 0.74-0.75$) in obese women. The relationship was weaker $(r=0.46)$ in obese men for whom waist circumference was not significant [14, 24, 35], suggesting that the predictive power of DXA combined with anthropometry to estimate IAF may be dependent on sex, and the degree of obesity in adult populations [35]. In older adults, both regional (trunk and manually defined ROI) and total abdominal fat masses from DXA were significantly correlated with VAT [34]. However, neither of these DXA measurements was superior to anthropometric measurements (waist circumference, sagittal diameter; r<0.74) [34], and models improved only slightly when combinations of DXA with anthropometry were examined.

To our knowledge, this is the first study to examine the associations of DXA android fat mass and anthropometry with VAT in adolescents and young adults. Despite the high intercorrelation between DXA trunk fat and android and L1-L4 regions (all r=0.97) in our study, trunk fat showed lower correlations with VAT ($r=0.68$) compared to AFM ($r=0.76$) and L1-L4 (r=0.71). Thus, android and regional L1-L4 fat masses were used as predictors because they are more anatomically associated with VAT. Importantly, L1-L4 has been validated in adults (20, 33, 53), to accurately estimate IAAT by CT, another reference standard for measuring visceral adiposity, and in adolescents (46) to predict metabolic risk factors associated with the accumulation of VAT.

Differences in the distribution of adipose tissue by gender are apparent as early as prepuberty, and the magnitude of the sex difference increases with maturation, with young adult males displaying higher relative central fat deposition and young adult females displaying more peripheral fat distribution compared with those in late adolescence [31]. Ethnic differences in abdominal fat distribution are also evident in young adults [32] and children [30], especially between Asian, Caucasian and African-American children [17, 36]. Ethnicity was not a significant predictor of VAT in this study, most likely because the number of subjects in each ethnic group was limited. Although DXA is considered a criterion method for assessing body composition, limitations in the use of DXA to predict VAT are evident from reports indicating that DXA significantly underestimates abdominal adiposity in individuals with less abdominal fat [37, 38] and overestimates this measure in individuals who are more obese or who have larger amounts of abdominal fat mass [13, 38– 42]. Earlier studies that investigated this issue showed that errors in estimates of body fat were positively correlated to tissue thickness [40]. Typically, the thicker the tissue under analysis, the more difficult it is for DXA to accommodate beam hardening at a preferential energy value and differentiate soft tissue composition. Tissue thickness >20 cm is projected to result in DXA overestimations of tissue fat [40, 43]. Because estimations in heavier individuals are subject to greater error that may introduce bias into regression equations that

predict VAT, population- specific (ie fatness groups) equations may be necessary to accurately predict VAT in overweight and obese individuals.

By its design, DXA cannot distinguish between intra-abdominal (IAAT) and subcutaneous (SAT) fat depots [36]. Several studies have investigated the utility of anthropometric variables (eg skinfold, abdominal thickness) to quantify regional adiposity (ie trunk, abdominal, gynoid) because of their practicality, accessibility, low cost, and reproducibility in the clinical setting [44]. Because android fat and the L1-L4 fat mass regions include VAT and SAT depots, using skinfold measurements to act as a surrogate of abdominal SAT [45] in combination with DXA abdominal fat measures may be beneficial in improving the accuracy of predicting abdominal VAT. Indeed previous studies have shown the use of skinfolds do improve the explained variance in models predicting IAAT by CT [1, 17].

Notably, because manually drawn ROIs are necessary to analyze L1-L4 regions, the potential for human error increases. Also, the anatomic arrangement of the ribs and spine may limit the area (number of pixels) for estimating bone-free soft tissue by DXA and consequently lead to underestimations of the total fat mass within the abdominal and thoracic area [10, 40]. Identification of the L1-L4 ROI maybe confounded by the degree of adiposity in this area, thereby reducing the clarity of the images and increasing the potential for observer error in delineating specific regions, for example, the respective inter-vertebral spaces [10, 36], an observation that was noted in our study when evaluating obese and overweight subjects. In fact, incorrect placement of the intervertebral disk spaces on the image for ROI placement is reported as the most common operator-dependent error when taking measurements in the spinal or thoracic cavity [46]. Thus, for DXA ROIs to be used as predictors of VAT, correct numbering of lumbar vertebral levels and correct ROI placement is imperative when analyzing abdominal adiposity [36, 40]. Use of standard, validated equations employed in the manufacturers' automated protocol for estimating AFM may help explain why AFM predicted VAT better than the manually drawn L1-L4 ROI. Additional factors that can influence DXA ROI placement include incorrect posture, overlapping of upper limbs or placement of upper limbs behind the trunk, vertebral conditions (i.e., floating ribs), and technical skill.

In summary, DXA AFM and L1-L4 ROI provide acceptable estimates of VAT in adolescents and young adults. Estimation was improved with the inclusion of gender and weight in models with AFM. Because gender appeared to be a moderator in the prediction of VAT, particularly with DXA L1-L4, the utility of different DXA ROIs to predict VAT may be dependent on gender, an issue that needs investigation. AFM was a better predictor of VAT than the manually drawn L1-L4 ROI, although the difference was not large. We conclude the combination of DXA-derived fat mass in the L1-L4 or android regions of interest with anthropometric measures (ie weight) can provide researchers and clinicians with a feasible, cost-effective and accurate method of estimating visceral adipose tissue.

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Abreviations

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Table 1

Descriptive characteristics by gender and of the total sample. Descriptive characteristics by gender and of the total sample.

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Bivariate correlations between covariates and fat masses. Bivariate correlations between covariates and fat masses.

I Results from bivariate correlations Fischer's transformation z-test concluded no significant differences in Pearson's correlation coefficients among male and females; thus, bivariate correlations are *Res*ults from bivariate correlations Fischer's transformation z-test concluded no significant differences in Pearson's correlation coefficients among male and females; thus, bivariate correlations are reported for the total sample. reported for the total sample.

Table 3

Multiple regression equations for estimating VAT from AFM.

VAT, visceral adipose tissue; AFM, android fat mass; Gen, gender; WT, weight

^{*} significant at *P* 0.05

Table 4

Multiple regression equations for estimating VAT from L1-L4 ROI.

L1-4FM, L1-L4 fat mass; Gen, gender

^{*} significant at *P* 0.05