Weight-loss diet alone or combined with resistance training induces different regional visceral fat changes in obese women

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Background: Quantification of abdominal fat and its regional distribution has become increasingly important in assessing the cardiovascular risk.

Objective: To examine the effects of 16 weeks of a hypocaloric diet with a caloric restriction of 500 Kcal per day (WL) or the same dietary intervention plus resistance training (WL + RT) on regional variation of abdominal visceral (visceral adipose tissue (VAT)) and subcutaneous (subcutaneous adipose tissue (SAT)) fat loss. Second, to identify the single-image that best represents total magnetic resonance imaging measurements of total VAT and SAT volume before and after WL or WL + RT intervention.

Design: A total of 34 obese (body mass index: 30–40 kg m⁻²) women, aged 40–60 years, were randomized to three groups: a control group (C; n = 9), a diet group (WL; n = 12) and a diet plus resistance training group (WL + RT; n = 13) with the same caloric restriction as group WL and a 16-week supervised whole-body RT of two sessions per week.

Results: WL + RT programs lead to significant changes in the location of highest mean VAT area from L3-L4 to L2-L3 discal level from pre- to post- intervention, whereas after WL the greatest relative VAT losses were located at L5-S1. Similar decreases in the SAT areas at all discal levels were observed after WL and WL + RT.

Conclusion: Different weight loss regimes may lead to different distribution of VAT. Sites located significantly above (cranial to) L4-L5 (that is, ~5–6 cm above L4-L5 or at L2-L3 discal level) provided superior prediction of total abdominal VAT volume, whereas more caudal slices provide better prediction of subcutaneous fat, not only before but also after either WL or WL + RT. International Journal of Obesity advance online publication, 7 September 2010; doi:10.1038/ijo.2010.190

Keywords: visceral adipose tissue; subcutaneous adipose tissue; exercise; MRI

Introduction

Bodyweight loss induced by either dietary intervention, alone or in combination with exercise (that is, aerobic or resistance training) lead to decreases in total visceral depot. These changes in abdominal fat and in its regional distribution have also been accompanied by significant improvements in different cardiovascular risk factors (for example, insulin sensitivity, baseline insulin levels, total cholesterol or low-density lipoprotein-C).¹⁻¹³ Magnetic resonance imaging (MRI) has been extensively used to measure adipose tissue-system level components in vivo.¹³⁻²² Limited by scanning time and the relatively high cost of image analyses obtained, several studies have attempted to precisely identify the best single-image location to best represent total volume of visceral adipose tissue (VAT) and subcutaneous adipose tissue (SAT). Previous epidemiologic and intervention studies have commonly used the L4-L5 image to examine whole-body MRI visceral fat measurement.²¹⁻²₅ Other studies, however, have considered the L2-L3 site and/or images located ~5–10 cm above the L4-L5 image to be more appropriate as they have been very highly correlated with total VAT volume.²₆⁻₂₈,²₉⁻₃₂ However, exercise program (that is, aerobic or resistance training), hypocaloric diet and/or combined interventions may lead to differential regional alterations of adipose tissue metabolism and regional VAT depot loss, possibly by mobilizing free fatty acids from VAT at different regions of the abdomen. Whether or not dietary intervention or a combined diet and resistance training changes the optimum anatomic site to prediction total VAT volume is a hypothesis that should be taken into consideration. Therefore, quantification of the degree of intra-subject regional variation in intra-abdominal

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VAT and SAT content (from xiphoid to symphysis using contiguous image data without gaps) need further attention. In addition, the ability to precisely identify the single-image that best represents total MRI measurements of VAT and SAT volume within the abdominal region in response to different interventions (that is, diet or diet combined with resistance training) has not been extensively examined.

The purpose of this study was to examine the effects of either 16 weeks of a hypocaloric diet with a caloric restriction of 500 Kcal per day (WL) or the same dietary intervention plus resistance training (WL + RT) on regional variation of abdominal VAT and SAT loss. A second objective was to compare the ability to precisely identify the single-image that best represents total MRI measurements of total VAT and SAT volume within the abdominal region following either a location relative to L4-L5 method, and a discal level approach before and after a diet or combined diet and resistance training intervention. We hypothesized differential effects on abdominal tissue distribution pattern of VAT and SAT depots depending on the diet and/or resistance training weight-loss intervention performed. Furthermore, after either WL or WL + RT interventions the ability to predict total VAT and SAT mass using single-slices images will be modified.

Methods

Subjects

A total of 34 sedentary, non-smoking, obese (body mass index (BMI): 30–40 Kgm$^{-2}$) women, aged 40–60 years, were recruited through an advertisement in a local newspaper. Before inclusion in the study all candidates were thoroughly screened using an extensive medical history, resting and maximal exercise electrocardiogram and blood pressure measurements. Subjects with cardiovascular, neuromuscular, arthritic, pulmonary or other debilitating diseases were excluded from the study. None of the subjects received any medication. All the subjects were informed in detail about the possible risks and benefits of the project, and signed a written consent form before participating in the study. This project was approved by the ethical committee of the regional health department. Participants were randomized to three groups: a control group (C; n = 9); a diet group (WL; n = 12) with a caloric restriction of 500 Kcal per day; and a diet plus resistance training group (WL + RT; n = 13) with the same caloric restriction as group WL and a 16-week supervised whole-body resistance training program of two sessions per week. Peri-menopausal women were balanced among groups. The subjects were tested on two different occasions (weeks 0 and 16) using identical protocols. During the 16 weeks of the study the subjects maintained their customary recreational physical activities (for example, walking). The baseline characteristics of the subjects are presented in Table 1.

MRI acquisition and image analysis

The MRI of the abdomen was performed with a 1Tesla magnet (Magnetom Impact Expert, SIEMENS, Erlangen, Germany) using a body coil. The subjects were examined in supine position with arms positioned parallel along the lateral sides of the body. The whole body fat was imaged using a ‘multi station’ approach at two different table positions within 5–10 min. We obtained a spoiled T1-weighted gradient-echo sequence; imaging parameters included field of view = 500 mm, matrix = 512 × 128, acquired in-plane resolution = 3.90 × 1.97 mm, reconstructed slice thickness = 10 mm (acquired: 10 mm), repetition time/echo time = 127/6 ms and number of slices = 10. Because of the limitation of the table-top translation, the subjects were imaged in two half-body volumes. Sagittal, coronal and transverse localizers of the abdomen from the diaphragm to the symphysis pubis were obtained to determine the precise anatomical sites for image acquisition, allowing location of each image to discal reference. Each half-body volume was scanned using two stacks, each containing 10 contiguous 10 mm-thick slices. Each stack was acquired in 20 s and interleaved slice order was used. All the stacks were acquired with breath holding during expiration. The total research time was about 5–10 min.

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The segmentation of SAT and VAT was performed by the same experienced operator. VAT was defined as adipose tissue, including the intra-abdominal cavity, region enclosed by the inner aspect of the abdominal wall and the anterior margin of the vertebral body. SAT was defined as the area enclosed by the innermost aspect of the abdominal muscle wall and the skin surface. Total abdominal fat (TAT) was defined as the summation of VAT and SAT areas. The acquired axial MR images were transferred to an external personal computer running Windows XP. We used a specially designed image analysis software (SliceOmatic 4.3, TomoVision, Montreal, QC, Canada) for quantitative analysis of the images. The model used to segment the various tissues is fully described and illustrated elsewhere. We used a region-based thresholding method (that is, ‘region growing’ function) to segment the fat regions. This method is based on the property of our acquired MR images in which the signal intensities of fat pixels at T1 images are higher than those of muscle and visceral organ pixels. Briefly, a multi-step procedure was used to identify tissue area (cm$^2$) for a...
| Table 1 Baseline and follow-up anthropometric characteristics pre and post 16 weeks of intervention (week 0 and week 16) |
|---|---|---|
| **C group (n = 9)** | **WL group (n = 12)** | **WL+RT group (n = 13)** |
| **Age (years)** | **51.5 ± 7.2** | **51.6 ± 6.6** | **47.7 ± 6.5** |
| **Antropometry** |  |  |  |
| **Weight (kg)** | 88.68 ± 12.7 | 88.15 ± 11.74 | 87.22 ± 18.01 | 80.81 ± 16.64* | 88.08 ± 12.63 | 80.36 ± 9.11*** |
| **BMI (kg m⁻²)** | 35 ± 3.6 | 35 ± 3.3 | 34.6 ± 3.4 | 32.4 ± 3.7*** | 35 ± 3.1 | 32.3 ± 3*** |
| **Waist (cm)** | 100 ± 7.4 | 99.9 ± 8 | 101.1 ± 6.5 | 94.5 ± 9.9*** | 99.7 ± 8.3 | 93.4 ± 8.8*** |
| **WHR** | 0.9 ± 0.05 | 0.9 ± 0.03 | 0.9 ± 0.03 | 0.9 ± 0.02** | 0.9 ± 0.03 | 0.9 ± 0.02** |
| **Abdominal MRI volume** |  |  |  |
| **VAT (cm³)** | 3370.78 ± 1228.4 | 3329.26 ± 1187.2 | 3243.75 ± 1085.3 | 2557.51 ± 1171.2*** | 3211.30 ± 1232.1 | 2528.43 ± 1039.9*** |
| **SAT (cm³)** | 14 225.21 ± 3347.6 | 14 021.24 ± 3127.2 | 13 340.57 ± 3411.1 | 11 204.76 ± 3326.8*** | 15 093.78 ± 2644.5 | 12 343.66 ± 2318.1*** |
| **TAT (cm³)** | 17 399.81 ± 3735.2 | 17 180.85 ± 3455.9 | 16 672.07 ± 3889.1 | 13 819.75 ± 4078.9*** | 18 327.64 ± 3035.5 | 14 921.62 ± 2821.1*** |
| **Baseline metabolic variables** |  |  |  |
| **Fasting plasma glucose (mg dl⁻¹)** | 98.4 ± 9.8 | 100.7 ± 11.2 | 98.6 ± 14.9 |
| **Insulin (mcU ml⁻¹)** | 16.8 ± 5.5 | 16.7 ± 9.2 | 15.1 ± 6.9 |
| **HOMA index (10⁻⁴ x μU⁻¹ x ml⁻¹)** | 4.2 ± 1.4 | 4.2 ± 2.4 | 3.7 ± 1.9 |
| **Baseline lipoprotein profiles** |  |  |  |
| **Triglycerides (mg per 100 ml)** | 140.2 ± 61.3 | 140.8 ± 48 | 111.2 ± 27.8 |
| **TC (mg per 100 ml)** | 262.6 ± 35.3 | 250.1 ± 42.4 | 248.6 ± 33.5 |
| **LDL (mg per 100 ml)** | 155 ± 29 | 147.8 ± 30.1 | 143.3 ± 27.6 |
| **HDL (mg per 100 ml)** | 64.6 ± 9.9 | 63.7 ± 14.5 | 69.3 ± 8.6 |
| **VLDL (mg per 100 ml)** | 28.1 ± 12.4 | 28.1 ± 9.5 | 22.2 ± 5.4 |
| **TC/HDL (ratio)** | 4.2 ± 0.8 | 4.1 ± 0.9 | 3.6 ± 0.6 |

Abbreviations: BMI, body mass index; HDL, high-density lipoprotein; HOMA, homeostasis model assessment; LDL, low-density lipoprotein; MRI, magnetic resonance imaging; SAT, subcutaneous adipose tissue; TAT, total adipose tissue; TC, total cholesterol; VAT, visceral adipose tissue; VLDL, very low-density lipoprotein; WHR, waist-to-hip-ratio. Values are expressed as means ± s.d. Differences between groups after intervention: *P < 0.05, **P < 0.01, ***P < 0.001 between values in weeks 0 and 16.
given MRI image. In the first step, the observer labelled the different tissues by assigning them different codes on the basis of each image’s histogram illustrating separate peaks for adipose tissue and lean tissue. The determination of an adequate threshold of the signal intensity was performed in each image individually to distinguish between adipose and lean tissues on MR images. Within the ‘Region Growing/ Painting’ control panel, the upper threshold was set to the maximum possible setting and the lower threshold was set to a value within the range of 300–650, with the exact value dependent on the pixel intensity of the SAT and VAT in the particular series of images, referred to the histogram. A region of interest describing the VAT area were created. Each consecutive slice was tagged using the same region growing procedure. The computer program connected and grouped automatically the pixels of similar signal intensities at VAT and SAT areas created by the regions of interest. At the second step, SAT and VAT tags resulting from the ‘Region Growing/ Painting’ mode were reviewed by an interactive slice-editor program, which allowed for verification and, where necessary, correction of the segmented results. The original gray-level image was superimposed on the segmented (for example, colour coded) image using a transparency mode to facilitate the corrections and edited as needed in the ‘Edit’ mode if image artefacts are identified, and to manually tag adipose tissue not previously tagged (adipose tissue areas below the chosen threshold). High-intensity non-fat pixels arising from bone marrow, liver and fatty intestinal contents within the VAT were avoided or manually removed. The area (mm²) of SAT and VAT in each image was computed automatically by summing the AT pixels and multiplying by the individual pixel surface. Summation of all the 1-cm slices provided total fat volume.

The discal level analysis was conducting labelling each image referred to discal spaces using sagittal scout images. Image related to L4-L5 analysis was conducted after labelled each image referred to L4-L4 discal level. Intra-observer reliability for calculation of total VAT, SAT and TAT volumes was of 0.99 with a coefficient of variation of 5–8%.

**Hypocaloric diet**

Each subject in WL and WL + RT groups received a varied and well-balanced hypocaloric diet (55% of calories as carbohydrates, 15% as proteins and the rest as fat) of 500 Kcal per day, according to the previous analysis of individual daily energy expenditure by accelerometry. This diet was designed to elicit a 0.5 kg weight loss per week. The control group was asked to maintain body weight. Through-out the 16-week intervention period body weight was recorded once every 2 weeks in both WL and WL + RT groups. Also, every 2 weeks each subject in the intervention groups participated in a series of 1-hour seminars in which the dietician taught proper food selection and preparation, eating behaviour, control of portion sizes and modification of binge eating and other adverse habits.

**Strength testing and training protocol**

The strength training program used in the present study was similar to that reported previously. Briefly, the subjects were asked to report to the training facility twice a week to perform dynamic resistance exercise for 45–60 min per session. A minimum of 2 days elapsed between two consecutive training sessions.

Each training session included two exercises for the leg extensor muscles (bilateral leg press and bilateral knee extension exercises), one exercise for the arm extensor muscle (the bench-press) and four to five exercises for the main muscle groups of the body. Only resistance machines (Technogym, Gambettola, Italy) were used throughout the training period. Resistance in this study was progressively increased or decreased every week for the 16-week training period using a repetition maximum approach, so that the loads that brought about a given relative intensity remained unchanged from week to week.

During the first 8 weeks of the training period the subjects trained with loads of 50–70% of the individual 1-RM (repetition maximum), and during the last 8 weeks of the training period the loads were 70–80% of the maximum. In addition, from week 8 to week 16 the subjects performed a part (20%) of the leg extensor and bench-press sets with loads ranging from 30 to 50% of the maximum. In all the individual exercise sessions performed one of the researchers was present to direct and assist each subject towards performing the appropriate work rates and loads. For all subjects average compliance with the diet classes and exercise sessions was above 95%.

**Statistical tests**

Standard statistical methods were used for the calculation of the means, s.d. One-way analysis of variance was used to determine any differences between the three groups’ initial measurements. The resistance training and/or diet-related effects were assessed using a two-way analysis of variance with repeated measures (groups × time). When a significant F-value was achieved, Bonferroni post hoc procedures were performed to locate the pairwise differences between the means. Selected relative changes were analyzed by one-way analysis of variance. Analyses of covariance were used to adjust post-interventional values to compare the data between the groups. For this purpose, pre-interventional values were used as covariates so that the effects of covariance could be observed. Pearson product–moment correlation was applied to identify the relationship among total VAT and SAT volumes and individual SAT and VAT images, areas calculated from each MRI method (that is, relative to discal level and to L4-L5 MRI methods). In addition, bivariate correlation was also applied to identify the relationship between magnitude of changes of total VAT and SAT volumes and magnitude of changes of single-image areas relative to discal level and to L4-L5 MRI methods as a result of WL or WL + RT. To test the similarity of slopes and intercepts of these relationships, the corresponding Student’s
Results

Baseline characteristics were similar in all groups. After 16 weeks, no significant changes were observed in the different parameters evaluated in the control group (Table 1). No significant differences were observed between WL and WL+RT groups in the magnitude of decrease in body weight, BMI and waist-to-hip-ratio after the 16 weeks intervention (Table 1).

*Abdominal adipose tissue distribution after WL or WL+RT*

At baseline, relative abdominal fat distribution for the whole group of patients was 18.82% for total VAT and 81.18% for total SAT volumes. After intervention, no significant differences were observed in relative abdominal VAT and SAT distribution after WL or WL+RT interventions. Statistical power calculations for this study ranged from 0.75 to 0.80. The P<0.05 criterion was used to ascertain statistical significance. SPSS (version 11.0, SPSS, Chicago, IL, USA) were used for descriptive statistics and analyses.

*Abdominal fat changes following each MRI analyze method*

**Image level relative to discal level method.** Total TAT, VAT and SAT were significantly reduced for WL and WL+RT groups (Figure 2). No significant differences were observed in the magnitude of decrease between WL and WL+RT groups in total VAT, SAT and TAT volumes. After intervention, VAT and SAT areas significantly decreased in most discal levels analyzed (P<0.05) except for VAT at L2-L3 in WL group, and for VAT at L5-S1 in WL+RT group. The magnitude of decrease in VAT at L5-S1 level was significantly greater in WL (24.8 %) compared with that observed in WL+RT (14.84%) (Figure 3). The magnitude of decrease in VAT at L2-L3 level was greater in WL+RT (22.36%) than that observed in WL group (18.05%). No significant differences were observed in the magnitude of decrease between WL and WL+RT in the SAT volumes at all discal levels analyzed.

**Image level relative to L4-L5 method.** After intervention, VAT volume decreased significantly (P<0.05) in most images levels referred to L4-L5 in WL group (except for L4-L5 + 8 cm, L4-L5 + 7 cm, L4-L5 + 3 cm, L4-L5 – 4 cm, L4-L5 – 5 cm, L4-L5 – 6 cm and L4-L5 – 9 cm image levels) and WL+RT (17.23 and 82.77%), respectively. Before intervention, the single image with the highest mean SAT area relative to discal level method was located at L5-S1 discal level and at 3 cm below L4-L5 discal level. As a result of WL or WL+RT, no significant differences were observed in the location of the single image with the highest mean SAT area with any of the MRI analysis methods performed compared with those reported before intervention. For VAT values the overall pattern was ‘slightly’ reversed from that observed for SAT (Figure 1). Before intervention, the single image with the highest mean VAT area was located at L3-L4 discal level and at 4 cm above L4-L5 for the whole group of subjects. As a result of WL, no significant differences were observed in the location of the highest single image with the highest mean VAT area within the abdomen with any of the MRI analysis methods performed. As a result of WL+RT, the location of the highest mean VAT area within the abdomen differ (P<0.01) to that reported before intervention. Thus, the highest single-image VAT location reached L2-L3 and 8 cm above L4-L5 discal level, respectively. The general pattern for TAT value distribution was similar to that observed for SAT. The single image with the highest mean TAT area within the abdomen was located at L5-S1 discal level and at 16 cm below L4-L5. As a result of WL or WL+RT, no significant differences were observed in the location of the single image with the highest mean TAT area.

**Relationships between total VAT and SAT volumes and single image areas relative to discal level and image location relative to L4-L5 MRI method before and after WL or WL+RT interventions**

**Discal level method.** The variation in correlations by discal level location for SAT and VAT are illustrated in Figure 5. Before intervention, the correlation between total SAT and VAT volumes and individual discal levels was moderate (generally r>0.81), whereas these were improved as a result of WL (r>0.92) or WL+RT (r>0.85). Before intervention, the discal level with the highest correlation with total SAT volume was located at L3-L4 discal level (r=0.92). After 16 weeks of intervention the corresponding correlation as a result of WL was changed to L5-S1 discal level (r=0.96), whereas as a result of WL+RT was located at L4-L5 (r=0.93) discal level. Before intervention, the discal level with the highest correlation with total VAT volume was located at L2-L3 discal level (r=0.90). As a result WL or WL+RT intervention, no significant changes were observed in the location of the discal level area with the highest correlation with total VAT volume (for example, at L2-L3; r=0.96 and r=0.96, respectively).
Before intervention, the correlation between total SAT and VAT volume and individual image areas relative to L4-L5 was moderate (generally $r \geq 0.74$), whereas these improved as a result of WL ($r = 0.93$) or WL + RT ($r = 0.81$). In contrast, the corresponding correlation coefficient for SAT volume decreased ($r = 0.79$), compared with previous intervention.

Relationships between magnitude of changes of total VAT and SAT volumes and magnitude of changes of single-image areas, relative to discal level and to L4-L5 MRI methods as a result of WL or WL + RT

Discal level method. Correlation coefficients between individual changes of total abdominal SAT volume and individual changes of single-slice changes in relation to the discal level method were generally high (mean $r = 0.83$) with a regional variation after WL (from $r = 0.73$ to $r = 0.93$), but with greater relative variability as a result of WL + RT (from $r = 0.63$ to $r = 0.91$) (Figure 6). The corresponding correlation coefficients for changes of total abdominal VAT volume and individual single image area changes in relation to the discal level method were lower than those observed for SAT as a result of WL (mean $r = 0.62$) and WL + RT (mean $r = 0.59$). Regional variations were also observed in both intervention groups so that the highest correlation coefficient between individual abdominal VAT area changes at different discal level slices and corresponding individual global VAT volume changes was located at L2-L3 as a result of WL ($r = 0.79$) and WL + RT ($r = 0.76$).

Location of image relative to L4-L5 method. The correlation coefficients between individual changes in total abdominal SAT volume and the corresponding individual changes of

Figure 1 Left: sagittal scout image; middle: distribution of cross-sectional area (mm$^2$) from xiphoid process (top) to symphisis pubis (bottom); right: cross sectional images at L2-L3 and L4-L5. The scout and the cross sectional images corresponded to a woman before (a) and after (b) WL + RT intervention. The distribution of cross-sectional area corresponds to the mean values of WL + RT group; in green VAT areas, in red SAT areas. We re-examined the sagittal scout images in all the subjects to determine the anatomical location of the image levels related to L4-L5 in reference to dorsolumbar vertebrae. The L4-L5 + 7 crossed the L2-L3 discal space in 54.5% of patients ($n = 18$).
**Figure 2** Absolute volumes of visceral fat (VAT), subcutaneous fat (SAT) and total abdominal fat (TAT) before and after control, diet or diet + resistance training interventions. *Significant differences (P < 0.05) compared with the corresponding baseline point (before vs after); †significant differences (P < 0.05) compared with the corresponding relative magnitude of change of the control group.

**Figure 3** Absolute volumes of visceral fat (VAT) and subcutaneous fat (SAT) within the abdominal region following a discal level approach before and after WL (a) or WL + RT (b) interventions. *Significant differences (P < 0.05) compared with the corresponding baseline point (before vs after); †significant differences (P < 0.05) compared with the corresponding relative magnitude of change in the control group. ‡Significant differences (P < 0.05) compared with the corresponding relative magnitude of change of the WL group.
single-slice areas in relation to L4-L5 were generally high as a result of WL (mean $r = 0.84$), but lower as a result of WL + RT (mean $r = 0.62$) (Figure 7). The corresponding correlation coefficients for individual changes in total abdominal VAT volume and single-slice area changes relative to L4-L5 were lower than those observed for SAT as a result of WL (mean $r = 0.69$) and WL + RT (mean $r = 0.51$) (Figure 7).

Further, highlighting the difference between the methods, the Bland-Altman plots demonstrate that there are a $\pm 4.4$ and $5.8\%$ coefficients of variance between log of the difference between total abdominal VAT and L2-L3 slice changes and changes in total abdominal VAT volume with a total error (log) of 8.8 cm$^3$ and 10.6 cm$^3$ as a result of WL and WL + RT, respectively (Figure 8).

Discussion

The findings of the present study show that either a diet or a combined diet and resistance 16-week training intervention lead to similar relative decreases in MRI measurements of total VAT, SAT and TAT volumes. However one of the key findings in the present study was that the abdominal tissue distribution pattern after WL differs to that observed after WL + RT. Therefore, our results show that 1) the WL + RT programs lead to significant changes in the location of highest mean VAT area from L3-L4 to that of L2-L3 discal level from PRE to POST intervention, whereas the overall VAT distribution pattern was not altered as a result of WL and 2) after WL the relative VAT loss at L5-S1 was greater than that in WL + RT and differed to the greater relative VAT loss observed at L2-L3 as a result of WL + RT. In contrast, to the differences in regional variations in VAT areas, the location of the highest mean SAT (that is, at L5-S1) before and after intervention, as well as the magnitudes of decrease in the SAT areas at all discal levels were similar in both WL and WL + RT. Also this is the first study to demonstrate that pre-intervention (that is, without any diet or exercise training effect), the relationship between total VAT and SAT volumes was moderate (generally $r > 0.81$), whereas the ability to predict VAT and SAT mass using single images was improved after both WL and WL + RT intervention as compared with subjects at baseline with overall higher BMI.
Before and after either WL or WL + RT interventions the single-slice VAT area with the highest correlation with either the corresponding individual total abdominal VAT volume or with the loss of abdominal VAT was located at L2-L3 discal level (that is, ~5–6 cm above L4-L5). Therefore, these findings suggest that different weight loss regimes may lead to different distribution of VAT. Sites located significantly above (cranial to) L4-L5 (that, ~5–6 cm above L4-L5 or at L2-L3 discal level) seem to be more suitable to predict total visceral fat, whereas more caudal slices provide better prediction of subcutaneous fat, not only before but also after either WL or WL + RT.

In line with previous studies, our data showed that 8 weeks intervention of a hypocaloric diet with a caloric restriction of 500 Kcal per day or a WL plus resistance training led to similar total VAT and SAT volume loss. This could be explained in part by the allometric relationship between changes of VAT and total fat mass during a weight-loss diet alone or combined with a resistance training program. Thus, Hallgreen and coworkers suggested that changes of VAT mass are determined primarily by fat-mass changes, as well as the initial ratio of VAT to fat-mass, and this is independent from weight-loss intervention. This relationship may explain the absence of differences in VAT and SAT volume reduction.

As seen in previous studies, however, our data also demonstrated at baseline (that is, before any diet or exercise intervention effect) regional variation in fat distribution in the whole group of subjects with the largest depot of absolute VAT and SAT at L3-L4 and L5-S1 discal level, respectively. Kanaley et al. showed that although the largest volume of abdominal fat was found in region at

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**Figure 5** Correlations between SAT, VAT and TAT total volumes and single image areas relative to discal level analyze method at baseline (a) and PRE and POST WL (b) or WL + RT (c) interventions.
L2-L3, this critical region had greater SAT, but smaller absolute VAT, than other regions located superiorly. The observation that ranking of subjects for VAT is influenced by measurement site has also been reported in previously premenopausal women\cite{28,30} and Caucasian men\cite{41}. However, a unique finding of the present study was that a weight-loss diet alone or combined with a resistance training program induce different regional changes in VAT volume, but not in SAT volume distribution patterns. Yet, after a combined weight-loss diet and resistance training intervention the location of the largest volume of VAT changed from L3-L4 to L2-L3 discal level, whereas the overall VAT distribution pattern was not altered as a result of a similar weight-loss diet alone. In contrast, to the regional variations in the overall VAT distribution pattern after \(\text{WL+RT}\), the location of the highest depot of absolute SAT remained at the same position (that is, at L5-S1 discal level) after both a weight-loss diet or \(\text{WL+RT}\) intervention. It is also likely that both intervention programs (that is, WL or \(\text{WL+RT}\)) induced a similar magnitude of SAT area decrease at all discal levels examined.

Taking our data as a whole, the present study showed differences in abdominal fat decreases depending on the diet and/or resistance training weight-loss intervention performed. This may suggest that WL alters adipose visceral tissue from L5-S1 abdominal level, whereas WL plus resistance training reduces this fat depot in higher scans of this region (for example, L2-L3), probably related in part to regional metabolic difference. This was true despite similar weight and waist circumference loss. Moreover, similar relative total VAT and SAT loss were obtained after both intervention programs. In agreement with our findings, Christiansen et al.\cite{36} demonstrated that 12-week randomized intervention led to comparable reduction of the VAT depot after a hypocaloric diet-induced weight loss (for example, 8 weeks of very low energy diet with a caloric restriction of 600 kcal per day, followed by 4 weeks of weight maintenance diet) with or without aerobic exercise in obese subjects. Similarly, Janssen et al.\cite{12} reported comparable reductions in VAT at L4-L5 after either a hypocaloric energy-restrictive diet (for example, to reduce their weight maintenance energy intake by 1,000 kcal per day with dietary fat intake restricted

Figure 6 Correlations between magnitudes of changes of total VAT, SAT and TAT volumes and corresponding magnitude of changes of single image areas relative to discal level method as a result of WL (a) or WL+RT (b) interventions.
to less than 30%) or the combination of a energy-restrictive diet with aerobic or resistance training on abdominal fat in obese women after a 16-week intervention period. Along these same lines, Redman et al.37 suggest that the changes in body composition and abdominal fat distribution are comparable after either 6 months of dietary caloric restriction (that is, 25% reduction in energy intake) only or dietary restriction in combination with endurance exercise. Kanaley et al.40 reported that after a 14-week weight loss intervention with diet and/or exercise the fat loss from the abdominal region is not uniform and greater relative VAT loss was found in the lower regions (that is, at L2-L3 and four slices below) of the abdomen than in the upper regions of the abdomen (that is, at four and eight slices above L2-L3).

Unfortunately, the differential effects on regional VAT loss of diet alone, exercise alone and diet plus resistance exercise were not compared separately in this study. Thus, resistance training had no additional effects on reduction of the VAT depot, compared with the major effects of hypocaloric diet alone. It is also likely that, by using the present resistance training program, energy expenditure was not substantial and would likely not have a significant additional impact on weight loss compared with WL intervention alone. However, a primary finding of the present study was that the effects of combined resistance training and a hypocaloric weight-loss diet per se on VAT does not seem to be uniform, with greater relative regional VAT loss in upper regions of the abdomen. Janssen et al.12 observed similar reductions in VAT and SAT fat depots in healthy men and women with either the combination of 16-week treatment of a combined diet and aerobic exercise, diet and resistance exercise or a hypocaloric diet with dietary fat intake restricted to less than 30% intervention alone. In contrast to our finding, Giannopoulou et al.10 demonstrated in women with type 2 diabetes that 14-week intervention of both a hypocaloric monounsaturated fat diet and a hypocaloric monounsaturated fat diet plus aerobic exercise reduced total and SAT abdominal fat, however, aerobic exercise is required for a reduction in VAT, because the hypocaloric monounsaturated fat diet alone did not reduce this fat depot. Thus, exercise program (that is, aerobic vs resistance training), hypocaloric diet and/or combined interventions may alter adipose tissue metabolism and regional VAT depot loss, possibly by mobilizing free fatty acids from VAT at different regions of the abdomen.

In agreement with previous studies,26–28,25,29–32 our results show that sites located significantly above (cranial to) L4-L5 (that is, L4-L5 +6 cm discal level or L2-L3 discal level) provided higher prediction of total abdominal VAT volume, compared with the association with VAT reported at L4-L5 discal level. A noteworthy finding of the present study was also that after both the intervention programs the measurement site located between L2-L3 discal level (or at ~5–6 cm above the L4-L5) provided even higher prediction of total VAT than observed at baseline level (that is, before WL or WL + RT). Therefore, this may suggest that quantification of intervention induced (that is, exercise and/or diet) abdominal total VAT changes may sub-estimate local abdominal fat changes at specific discal sites. Previous epidemiologic and intervention studies have commonly used the L4-L5 image to examine whole-body MRI visceral fat measurement.23–25 Several studies, however, have also considered more appropriate the L2-L3 site and/or images located ~5–10 cm above the L4-L5 image as they were very highly correlated with total VAT volume.26,42,27–30,12 An advantage of the present study was to examine the regional variation in intra-abdominal VAT and SAT content, as well as comparing the
ability to precisely identify the single-image that best represent total MRI measurements of VAT and SAT volume within the abdominal region following either a location relative to L4-L5 method and one following a discal level approach. In the present study, therefore, we used contiguous image data without gaps from xiphoid to symphysis allowing more precisely indentify the best single-image location to best represent total VAT and SAT volume. In doing so, we evaluated the degree of correspondence between multiple-image protocols to estimate abdominal VAT and SAT volumes performed in many previous studies. In addition, the present study examined separately the interventions effect of either a WL with a caloric restriction of 500 Kcal per day or a WL plus resistance training, which allowed us to compared the differential effects on regional variation of VAT and SAT distribution, as well as differences in abdominal region decreases in fat depending on the diet and/or resistance training weight-loss intervention performed. A limitation of the present study was that it included only a special clinical population (that is, perimenopausal obese women), so that our results should be applied cautiously in healthy adults of white,
black or Asian origin. It is also likely that the prediction of total VAT and SAT mass from single-slices maybe also compared with a group with similar weight and/or BMI to those reported after different weight loss regimes examined (that is, either a diet or a combined diet and resistance 16-wk training intervention).

In agreement with previous studies,28,29 our results also revealed that in pre-intervention the relationship between single-SAT areas and total SAT volume were uniformly high at all image locations with the highest relationship located at site measurement L3-L4 discal level \((r=0.92)\). In these instances, the ability to predict total SAT volume using single images after both WL or WL + RT intervention were improved, however, the location of site measurement changed to L5-S1 after WL and at L4-L5 discal level as a result of WL + RT. Therefore, at baseline status the optimum measurement site for prediction of SAT volume will be slightly different to that reported for VAT volume prediction, whereas it will change after WL or WL + RT interventions.

In conclusion, the findings of the present study show that, different weight-loss regimes may lead to different distribution of VAT. Thus, after a combined weight-loss diet and resistance training intervention the location of the largest volume of VAT changed from L3-L4 to L2-L3 discal level but, the overall VAT distribution pattern was not altered as a result of a similar weight-loss diet alone. The location of the highest depot of absolute SAT remained at the same position (that is, at L5-S1) after both WL or WL + RT. Sites located at L2-L3 discal level (that is, \(\sim 5-6\) cm above L4-L5) seem to be more suitable to predict total visceral fat, whereas more caudal slices provide better prediction of subcutaneous fat. In these instances, the ability to predict VAT and SAT mass using single images was improved after both WL or WL + RT as compared with subjects at baseline with overall higher BMI. This should be taken into consideration to accurately estimate SAT and VAT volume before and after diet, an exercise program or a combined diet and exercise program.35,44,45

Conflict of interest

The authors declare no conflict of interest.

References

Abdominal tissue distribution pattern after diet or combined with resistance training


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