Effects of energy restriction and exercise on skeletal muscle and adipose tissue in women as measured by magnetic resonance imaging\(^1\)\(^-\)\(^3\)

Robert Ross, Heather Pedwell, and John Rissanen

**ABSTRACT** The effects of energy restriction alone (diet alone, DO) and energy restriction combined with endurance exercise (diet and exercise, DE) on lean tissue (LT), skeletal muscle (SM), and adipose tissue (AT) were evaluated in 12 obese women (DO, \(n = 12\); DE, \(n = 12\)). The prescribed diet created an estimated daily deficit of 4.5 MJ (1000 kcal). LT, SM, and AT were measured by using magnetic resonance imaging (MRI). The weight loss observed for the DO group (\(-10.0 \pm 4.0 \text{ kg}\)) was not significantly different from that of the DE group (\(-11.7 \pm 3.0 \text{ kg}\)). However, the composition of the weight loss was significantly different between groups because the DE group lost significantly greater (\(P = 0.05\)) quantities of AT (\(-11.3 \pm 3.8 \text{ L}\)) than the DO group (\(-8.3 \pm 3.6 \text{ L}\)). Furthermore, whereas LT measured by MRI was maintained in the DE group, a modest (\(\approx 4\%\)) but significant (\(P < 0.01\)) reduction in LT was observed in the DO group. Similarly, measurement of SM by MRI in the appendicular region revealed a preservation of SM in the DE group, but a significant (\(P < 0.01\)) reduction in the DO group. Maximal oxygen consumption (\(L\)) was significantly improved (\(\approx 9\%\)) in the DE group (\(P < 0.01\)). These findings provide evidence that the combination of energy restriction and exercise result in a greater reduction of AT and preservation of LT and SM compared with energy restriction alone.

**KEY WORDS** Obesity, magnetic resonance imaging, adipose tissue, skeletal muscle, lean tissue, diet and exercise, weight loss

**INTRODUCTION** Whether the addition of physical activity to a regimen of energy restriction provides added benefits with respect to weight loss, adipose tissue (AT) reduction, and the preservation of lean tissue (LT), has been the subject of recent research and debate. At the present time a definitive answer to this question is not possible. For example, although it has been reported that exercise increases the rate of fat loss (1, 2), others have reported no increase (3). Similarly, exercise has been shown to maintain LT by some (4–6) but not by others (2, 7, 8). Although several explanations have been proposed that might explain the equivocal findings (9, 10), there is little consensus. Several investigators (10, 11) have argued that methodological problems related to the measurement of change in body composition may, in large measure, explain the equivocal nature of those studies that have attempted to describe the composition of diet- and exercise-induced weight loss. In most cases the method of choice for measuring change in body composition is hydrostatic weighing (HW) using the two-component model. Concerns about this method are well documented (9, 11, 12) and will not be repeated. However, the methodological problems associated with HW and other two-component models underscore the need for improved methods of measuring change in human body composition. Magnetic resonance imaging (MRI) is not subject to the inherent assumptions of two-component models and has been shown to provide accurate measures of both AT and skeletal muscle (SM) by comparison with human cadaver (13, 14) and animal data (15, 16). It has also been demonstrated that MRI can be used to measure exercise-induced changes in skeletal muscle (17).

The use of MRI offers two distinct advantages when assessing the influence of weight loss on body composition. The first is that regional effects, if any, of the given intervention can be evaluated. Thus, for example, it is possible to evaluate whether the increase of LT in one anatomical region is being masked by a loss of LT in another. The second advantage is that the use of MRI permits direct measurement of SM. Although we restrict our observations to measurement of SM in the appendicular region, this advantage is unique in that the effects of dietary or exercise-induced weight loss on SM per se have not been previously investigated.

In a previous investigation we used MRI to measure the combined effects of energy restriction and exercise on subcutaneous and visceral AT distribution (18). In this study we extend our previous investigation by comparing the combined effect of energy restriction and exercise with energy restriction alone on SM, LT, and AT in women.

**SUBJECTS AND METHODS**

**Subjects**

Inclusion criteria required that the women were premenopausal, were upper-body obese (body mass index (BMI, kg/m\(^2\))

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> 27 and waist-to-hip ratio (WHR), derived by using the umbilicus waist circumference > 0.85), were weight stable (± 2 kg) for 6 mo before the beginning of the study, were taking no medication known to affect the study variables (ie, oral contraceptives), and consumed on average less than two alcoholic beverages per day.

Initially 33 subjects participated in this study: 14 in the diet and aerobic-exercise group (DE) and 19 in the diet-only group (DO). For various reasons two women dropped out of the DE group and seven from the DO group before completion of the study. Twenty-four women complied with the study requirements, 12 in each group. The descriptive characteristics of both groups are given in Table 1. The two groups were not significantly different with respect to age, BMI, WHR, total LT, or total AT (P > 0.05). The study was conducted in accordance with the ethical guidelines of Queen's University.

Tissue measurement by MRI

Magnetic resonance images were obtained with a Siemens 1.5-T whole-body scanner (Erlangen, Germany). A T1-weighted, spin-echo sequence with a 210-ms repetition time and 15-ms echo time was used to acquire all the MRI data. The MR images in the abdomen region were obtained by using a rectangular field of view (192 × 256) and a one-half Fourier transformation sequence. Use of these parameters reduced the time required to obtain the data set in the abdominal region (seven images) from the ~8 min previously reported (19), to 26 s. During this 26-s period the subjects were asked to take a normal inspiration and hold their breath. Although the signal-to-noise ratio is decreased when this procedure is used, this limitation is offset by the reduction in respiratory motion artifact normally associated with the acquisition of MR images in the abdominal region. The total time required to obtain all MR data (41 images) for each subject was ~25 min.

The protocol used to acquire the MRI data is described in detail elsewhere (19). Briefly, the subjects lay in the magnet in a prone position, arms placed straight above the head. For all subjects six data sets (seven images for each set) were required to cover the entire body. Transverse slices (10-mm thickness) were acquired every 50 mm from head to toe by using the image at LA-LS as the point of origin. For all subjects a total of 41 images were acquired. All image data were transferred onto a stand-alone Personal Iris computer (Silicon Graphics Inc, Mountain View, CA) for analysis with software developed within our laboratory.

Calculation of lean, adipose, and skeletal muscle tissue area and volume

The model used to segment the various tissues is described elsewhere (19). The threshold selected for AT and LT was based on the analysis of a sample of typical images and their respective grey-level histograms. Once the appropriate threshold was determined the next step involved viewing the pixels that were identified as AT and LT in response to the threshold selected. Each slice was reviewed by using an interactive slice editor program that allowed for verification and, where necessary, correction of the segmentation result. The operation was facilitated by superimposing the original grey-level image on the binary segmented image using a transparency mode.

To calculate tissue area, the areas (cm²) of the respective tissue regions in each slice were computed automatically by summing the given tissues’ pixels and multiplying by the pixel surface area. The volume (cm³) of the respective tissues (ie, AT or LT) in each slice was calculated by multiplying the tissue area (cm²) by the slice thickness (10 mm). Whole-body AT and LT volumes were calculated by adding the volumes of truncated pyramids defined by pairs of consecutive slices (19). Whole-body volume was calculated by using all 41 slices. To determine whether regional differences existed with respect to the effects of either treatment on MRI-measured LT, the whole body was divided into upper (21 images) and lower (20 images) body regions with LA-LS used as the dividing point.

Because the pixel intensity values of many MRI-measured lean tissues fall within a small range (ie, organ and skeletal muscle), discrimination between them within the abdomen and torso regions is ambiguous. Therefore, measurements of SM by MRI are restricted to the arm and leg regions. Because MRI-measured LT in these regions comprises both SM and bone, the estimation of SM requires determination and subtraction of the bone area. Because the protons within the bone cortex have no appreciable MR signal, the cortex appears black on the image screen. Thus, by using a mouse pointer to delineate the borders of the cortex, the bone area in the arm and leg is determined in a straightforward manner. Arm SM volume (L) was calculated

<table>
<thead>
<tr>
<th>TABLE 1</th>
<th>Pre- and posttreatment anthropometric values†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Variable</td>
<td>Diet-only group</td>
</tr>
<tr>
<td></td>
<td>Pre</td>
</tr>
<tr>
<td>Age (y)</td>
<td>39.7 ± 6.8</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>91.2 ± 15.1</td>
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<tr>
<td>BMI (kg/m²)</td>
<td>33.7 ± 4.3</td>
</tr>
<tr>
<td>WHR²</td>
<td>0.84 ± 0.05</td>
</tr>
<tr>
<td>Circumferences (cm²)</td>
<td>—</td>
</tr>
<tr>
<td>Waist²</td>
<td>100.4 ± 12.9²</td>
</tr>
<tr>
<td>Upper arm</td>
<td>36.5 ± 2.9</td>
</tr>
<tr>
<td>Hip</td>
<td>119.7 ± 11.6</td>
</tr>
<tr>
<td>Proximal thigh</td>
<td>69.0 ± 3.6</td>
</tr>
</tbody>
</table>

† Data ± SD; n = 12 for both groups. Percent change in parentheses.
² Significantly different from pretreatment value, P < 0.05.
³ Waist-to-hip ratio derived by using the last-rib waist circumference.
⁴ Obtained at the level of the last rib.
by using the MR images extending from the hand to the humeral head. Thus, arm SM includes a large measure of the shoulder. In most cases the humeral head was visible; for those individuals where the humeral head was not visible, an estimation of the appropriate image was required. Leg SM was calculated by using the MR images extending from the foot to the femoral head, thus leg SM includes a large portion of the gluteal SM. Though not anatomically landmarked, the femoral head was visible for all subjects both pre- and posttreatment.

Reliability

Reliability of the measurements of LT by MRI was assessed by comparing duplicate (same day) MR images obtained on 19 of the subjects at the L4–L5 level. The correlation coefficient obtained between the two measurements was 0.94 (P < 0.0001). The difference between the two mean values was 1.0 ± 5.1% (P > 0.10, paired t test). The repeatability of whole-body LT volume (L) measurements was assessed from repeated measurements on two obese male subjects. For each subject a complete data set was acquired (41 images) on two separate occasions during the same day. The mean difference between tests 1 and 2 for MRI-measured LT (L) was < 2%. The MRI-measured LT calculations were determined by a single individual and thus represent the intrasubject error associated with repeated LT-volume calculations. In previous studies we reported that for total AT volume (L), the mean difference between tests 1 and 2 was 2.6% with a range of 0.9–4.3% (19). We also reported that the mean difference for repeated whole-body AT measurements in animals was < 4% (16).

Anthropometric variables

Body weight was measured on a balance scale calibrated to 0.1 kg. Barefoot standing height was measured to the nearest 0.1 cm by using a wall-mounted stadiometer. Circumference measurements were obtained with the subjects in a standing position at the following sites: biceps, forearm, chest, hip, proximal thigh, and calf and waist at the umbilicus (waist1) and last rib (waist2) levels by using the procedures described in the Anthropometric Standardization Reference Manual (20). Body fat distribution by anthropometry was estimated using the waist-to-hip ratio (WHR).

Diet and exercise regimens

Dietary protocol. Basal energy requirements were estimated by using the Harris-Benedict equation (21) multiplied by a factor of 1.5. It has been reported that the difference between the estimated and actual energy requirements in healthy subjects is < 8% (22) when this method is used. A weight-maintenance diet at the prescribed energy intake was followed for 1 wk. For 16 wk after the baseline period, the weight-maintenance energy intake was reduced by 4.19 MJ (1000 kcal/d). After completion of the 16-wk treatment period, the energy intake required to maintain weight was recalculated by deriving the average daily energy intake obtained from the diet records, and adding to that number the energy value associated with the weight loss (assuming 32.2 MJ/kg, or 3500 kcal/lb). The derived value was prescribed and followed for 1 wk. Subjects were required to limit the contribution of fat to the total energy intake to 30%. The foods consumed were self-selected; no supplements were prescribed. Food diaries were recorded daily for the duration of the 18-wk study, and were analyzed weekly to ensure adherence to the dietary protocol and to ensure that proper nutrition was maintained.

Aerobic exercise protocol. In addition to the energy-reduced diet, 12 women performed aerobic exercise 5 d/wk. The initial duration of exercise was ~ 15 min and progressed to a maximum duration of 60 min according to individual capabilities. Exercise intensity (heart rate) was monitored at each session by using an automated heart rate monitor (Polar USA, Inc, Stamford, CT). The mode of exercise was determined by the subjects and varied between stationary cycling, walking on a motorized treadmill, and stair stepping. All exercise sessions were by appointment and supervised by a physical educator.

Maximal oxygen uptake (VO₂ max) was determined by using a treadmill test that used a constant speed of either 4.8 kmh (3 mph) or 5.6 kmh (3.5 mph) depending on the subject's capabilities. The initial grade was set at 0% for the first 2 min, after which it increased by 2% for the third minute, and by 1% every minute thereafter. Standard spirometry techniques with a Beckman metabolic measurement cart (Sensormedics Inc, Fullerton, CA) were used to determine oxygen uptake. It was assumed that VO₂ max was attained when no increase in VO₂ was observed despite further increases in treadmill grade.

Energy cost of exercise

The oxygen cost of both treadmill walking and stationary cycling were determined by using the equations given by the American College of Sports Medicine (23). Howley et al (24) previously reported that direct measurement of MET (multiple of resting metabolic rate) values when a stair stepper was used were ~ 20% lower than those presented when the equation provided by the manufacturer was used. Therefore the MET values obtained when the stair stepper was used were reduced by 20% before estimating the oxygen cost. Energy expenditure of all three modes of exercise was subsequently determined by multiplying the oxygen cost by 21.1 kJ/L (5.04 kcal/L).

Statistical analysis

Data are presented as mean ± SD. Paired t tests using the absolute values obtained pre- and posttreatment were used to assess the within-group response to the respective treatments. Independent t tests using the absolute individual change scores (pre- minus posttreatment) were used to analyze between-group differences. Statistical procedures were performed by using SYSTAT (25).

RESULTS

With few exceptions, complete dietary intake records were submitted as required by all subjects. Analysis of the diet records indicated that the mean energy deficit created by the reduction in energy intake was 5.1 ± 1.2 MJ (1222 ± 293 kcal/d) for the DO group and 5.5 ± 1.1 MJ (1325 ± 253 kcal) for the DE group (P > 0.10). The mean dietary fat intake for the DO and DE groups was 21.5 ± 4.7% and 23.7 ± 4.9%, respectively (P > 0.05).

Attendance at the exercise sessions averaged 92% (range 85–98%) for the DE group. The duration of the exercise sessions averaged 34.0 ± 6.0 min, increasing from a mean of 20.1 ± 6.7 min in week 1 to a mean of 36.2 ± 5.8 min in week
16 ($P < 0.01$). The average exercise intensity for the 16-wk exercise period was $77.0 \pm 4.0\%$ of the maximum predicted heart rate (220 - age). With respect to exercise modality, 65% of the exercise was performed as stair stepping, 28% on the treadmill, and 7% on the stationary bicycle. Seven of the 12 DE group subjects performed > 75% of their exercise on the stair stepper. The mean energy expenditure for the DE group was $77.7 \pm 16.0$ MJ (18 562 \pm 381.5 kcal).

As part of the program the DO group was asked to attend weekly group meeting sessions intended to provide dietary counsel and develop individual success strategies. Adherence to these sessions averaged 85% (range 55–100%).

The pretreatment VO$_2$ max value for the DO group ($2.33 \pm 0.3$ L) was not significantly different from that of the DE group ($2.42 \pm 0.2$ L). In response to the aerobic exercise program peak VO$_2$ increased by 8.7% ($P < 0.01$) within the DE group (0.21 \pm 0.2 L). Within the DO group a 9.2% decrease in VO$_2$ was observed ($P < 0.01$).

The changes observed for selected anthropometric variables are given in Table 1. With the exception of WHR, significant reductions were observed for all anthropometric variables within both the DO and DE groups ($P < 0.01$). A between-group analysis revealed that the reductions observed for all variables were not significantly different.

The changes observed for total AT are given in Table 2. Within both groups significant reductions in total AT were observed ($P < 0.01$). A between-group analysis demonstrated that the reduction in AT for the DE group (11.3 \pm 3.8 L) was significantly greater than that observed for the DO (8.3 \pm 3.6 L) group ($P = 0.05$).

The changes observed for MRI-measured LT are given in Table 2; a significant ($P < 0.05$) reduction was observed for the DO group (− 1.5 ± 1.1 L) but not for the DE group. Within both groups this observation remained true for both upper- and lower-body regions. Thus, for the DO and DE groups, respectively, the loss and preservation of MRI-measured LT was uniform over the entire body. Note however that although statistically significant, the reduction in LT within the DO group was relatively small, ±3.5% (Table 2).

The evaluation of whether either treatment had effects on MRI-measured SM was restricted to SM in the appendicular region. As illustrated in Figure 1, MRI-measured SM volume was significantly reduced ($P < 0.01$) within the DO group for both the arm (arm and shoulder) and leg (buttocks and legs) regions, whereas no significant changes were observed in either region for the DE group.

The percent contributions of both AT and LT to the reduction in total (whole body) volume (L) are presented in Figure 2. For the DO group, AT represented 86% of the total volume reduction with LT representing 14%. For the DE group AT represented 98% and LT 2% of the reduction in total body volume. As a consequence significant increases ($P < 0.01$) in the ratio of LT to AT, derived by whole-body LT and AT measurements, were observed for both the DO (0.96 vs 1.13) and DE (0.95 vs 1.27) groups. A between-group comparison showed that the increase in the ratio of MRI-measured LT to AT observed for the DE group tended to be greater than that observed for the DO group ($P = 0.068$).

**DISCUSSION**

The principal finding of this study is that the combination of energy restriction and exercise resulted in a greater reduction of MRI-measured AT and preservation of MRI-measured LT compared with energy restriction alone. This, combined with the observation that functional capacity (peak VO$_2$) was significantly improved within the DE group, provides strong evidence in support of the combination of endurance exercise and moderate energy restriction as an efficacious means of treating obesity.

The findings reported here agree with those of a previous study in which we noted that the combination of energy restriction and resistance exercise resulted in the maintenance of MRI-measured LT and SM in a group of obese women (unpublished observations). Similar to the findings in that report, in this study we observed a preservation of LT in the upper- and lower-body regions that was not significantly different. Thus there was no compensatory increase in LT in one anatomic region that may have been masked by a loss of LT in another. Similarly, we observed that MRI-measured SM was maintained in both the arm (arm and shoulder) and leg (leg and gluteal) regions. On the basis of these findings we make two important observations. First, that the maintenance of MRI-measured LT in the DE group was uniform throughout the body, and second, that in this study the preservation of LT was synonymous with the maintenance of SM. The latter is a unique observation because we are unaware of other investi-

![Table 2](image)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Diet only group</th>
<th>Diet + exercise group</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pre</td>
<td>Post</td>
</tr>
<tr>
<td></td>
<td>Pre</td>
<td>Post</td>
</tr>
<tr>
<td>Lean tissue</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whole body (L)</td>
<td>$41.7 \pm 4.5$</td>
<td>$40.2 \pm 3.8$</td>
</tr>
<tr>
<td>Upper body (L)</td>
<td>$23.3 \pm 2.4$</td>
<td>$22.5 \pm 2.1$</td>
</tr>
<tr>
<td>Lower body (L)</td>
<td>$18.5 \pm 2.3$</td>
<td>$17.6 \pm 1.9$</td>
</tr>
<tr>
<td>Adipose tissue</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whole body (L)</td>
<td>$45.4 \pm 12.8$</td>
<td>$37.1 \pm 10.7$</td>
</tr>
</tbody>
</table>

1 $\bar{x} \pm SD; n = 12$ for both groups. Percent change in parentheses.

2 Significantly different from pretreatment value, $P < 0.05$.

3 Significantly different from DO group, $P < 0.05$. 
in free-living subjects is related to the degree of adiposity (28, 29), which suggests that the underreporting, if any, should have been similar between the two groups. It is also unlikely that the DE group consumed more food in response to exercise because obese, untrained subjects do not commonly alter their energy intake when exposed to exercise training (30).

Our finding that the combination of exercise and energy restriction preserved LT contrasts with the observations made in a recent review by Ballor and Pochiman (9), that a diet- and exercise-induced weight loss similar to that reported in this study (~10 kg), is associated with an ~12% decrease in LT for both men and women. Although several explanations have been suggested that may explain the equivocal findings (9, 10), the discrepancies may in large measure be explained by differences in the methodologies used to measure LT. Several investigators have argued that assumptions inherent to the use of a two-component, hydrostatic weighing model preclude its use as a reference measure of change in human body composition (9, 11). The principal concern lies in the inability of the method to measure change in total body water. Because total body water increases in response to a program of diet and exercise (31, 32), it has been argued that the use of a two-component hydrostatic weighing method, for example, will result in an overestimation of the preservation of LT, and consequently an underestimation of AT loss. This observation underscores the need to use more definitive techniques when measuring the effects of weight loss on body-composition indexes. MRI is not subject to the assumptions inherent to two-component models and has been shown to provide accurate measurements of AT and SM as compared with human-cadaver (13, 14) and animal data (15, 16). In addition, it has also been demonstrated that MRI can detect small changes in SM induced by exercise (17). Thus, although there is a need for data that would further validate the use of MRI as a measure of human body composition, at this point we are confident that the changes observed in MRI-measured LT and -AT reflect true biological changes.

The mechanism by which endurance exercise might induce a preservation of LT is unknown. Although direct stimulation of SM by endurance exercise is of an intensity below that thought to stimulate muscle hypertrophy (33), Goldberg et al. (33) reported that in animals, repeated muscle stimulation and/or simple passive stretching of muscle leads to a decreased rate of protein degradation postexercise. Thus, it is possible that the repetitive nature of endurance exercise contributes to the maintenance of LT by reducing the degradation of SM protein that occurs during periods of underfeeding. However, it is unlikely that this mechanism is entirely responsible for the observed preservation of SM because it fails to explain why SM was maintained in the arm and shoulder area, an anatomical region that was not routinely exercised by our subjects. Thus, rather than the direct stimulation of SM, it is more likely that the maintenance of LT was the indirect result of a preferential mobilization of fat as an energy source in the DE group. Consistent with this observation is the finding that within energy-restricted rats, the preservation of protein in SM is positively related to the degree of adiposity (34).

As with LT, the literature is equivocal as to whether the addition of exercise to energy restriction increases the rate of AT loss. For example, the combination of exercise and energy restriction have been shown to increase (1, 6) or have no effect

**FIGURE 1.** Comparison of pre- (■) and posttreatment (□) skeletal muscle tissue within the leg and arm regions for both the diet-only (DO) of diet + exercise (DE) groups. x ± SD. *P < 0.01.

**FIGURE 2.** Respective contribution of adipose tissue (AT, ■) and lean tissue (LT, □) to the reduction in whole-body volume for both the diet-only (DO, n = 12) and diet + exercise (DE, n = 12) groups.

REFERENCES

In summary, we demonstrated that the addition of endurance exercise to a program of energy restriction results in a reduction of MRI-measured AT and a preservation of both MRI-measured LT and SM which are greater than that induced by energy restriction alone. Combined with the observation that the exercise prescribed was well tolerated and induced a significant improvement in functional capacity, these findings suggest that this treatment is an efficacious means of reducing adiposity. There is a need for further study using an imaging method such as MRI or computed tomography to confirm these observations in male populations.


