Reduced physical activity increases intermuscular adipose tissue in healthy young adults[1–3]

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ABSTRACT

Background: Recent findings suggest that higher levels of intermuscular adipose tissue (IMAT) are associated with glucose dysregulation, lower levels of muscle strength, and a heightened risk of disability. Although several studies have described adaptations in muscle after reduced physical activity, the change in IMAT in healthy young adults is unknown.

Objective: The objective was to determine whether reduced lower limb activity alters IMAT in healthy young adults and to assess whether this change affects muscle strength loss.

Design: The subjects (6 men and 12 women aged 19–28 y) underwent a 4-wk control period, which was followed by 4 wk of unilateral lower limb suspension. Volumes of whole muscle, subcutaneous adipose tissue, and IMAT were assessed by using magnetic resonance imaging in the thigh and calf. Muscle strength was assessed during maximal voluntary isometric contractions.

Results: No changes were observed in the control period. Reduced physical activity decreased thigh and calf muscle volumes by 7.4% and 7.9% ($P < 0.001$), respectively; no significant change in subcutaneous adipose tissue was observed. Additionally, IMAT increased in both regions; the increase was larger in the calf (20%) than in the thigh (14.5%) ($P \leq 0.005$) and was partially explained by the loss in muscle ($R^2 = 26\%$). The loss in strength was greater in the thigh (20.4%) than in the calf (15%). Strength loss was associated with increases in IMAT ($P = 0.039$) after adjustment for the loss in muscle, initial strength, initial IMAT, and initial muscle volume.

Conclusions: IMAT accumulates markedly after reduced activity in healthy young adults. Increases in IMAT may contribute to losses in muscle strength associated with reduced physical activity, but the mechanism responsible is yet to be determined. *Am J Clin Nutr* 2007;85:377–84.

KEY WORDS Unilateral limb suspension, bed rest, physical inactivity, intermuscular adipose tissue

INTRODUCTION

Along with abdominal obesity and insulin resistance, a low level of physical activity is a major lifestyle risk factor for the development of the metabolic syndrome (1). The link between physical activity and factors that constitute the metabolic syndrome are partially mediated through skeletal muscle (2). Biological evidence suggests that reduced physical activity blunts key endothelial enzymes needed for triacylglycerol catabolism, which allows higher concentrations of triacylglycerols to accumulate in the blood (3). Furthermore, low levels of physical activity attenuate fatty acid oxidation in the muscle, which creates an environment for adipose tissue accumulation (4). However, it is not clear whether intermuscular adipose tissue (IMAT) increases after reduced physical activity in humans.

IMAT is only recently gaining attention as a potential contributor to glucose disposal and muscle function (5–7). For example, higher contents of adipose tissue or lipid in the muscle are reported in the hemiparetic leg after stroke and are associated with insulin resistance in lipodystrophic HIV-infected patients (8, 9), spinal cord–injured patients (10), and diabetic persons (6, 7). Furthermore, weight-loss and exercise interventions in diabetic persons reduced IMAT and positively shifted metabolic variables in a study y Ryan et al (11). A higher muscle lipid content, measured as muscle attenuation with computer tomography, is associated with lower levels of muscle strength and physical performance (5, 12). However, these studies were cross-sectional evaluations in aging or pathologic models; no longitudinal evaluations were made to empirically determine whether lower levels of physical activity induce changes in IMAT and consequently affect muscle strength.

Reductions in physical activity are well known to decrease muscle strength (13). This decrease in muscle strength was first thought to be a result of losses in muscle mass, but several studies suggest that changes in muscle mass are only weakly correlated with changes in muscle strength (14, 15). Changes in adipose tissue after reductions in activity have received less attention;

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however, because adipose tissue is well recognized as an endocrine organ, the cytokines released from accumulated IMAT could induce contractile damage (16–18). For example, higher cytokine concentrations, especially tumor necrosis factor α (TNF-α) (19), are associated with reductions in muscle mass and performance (20, 21). Therefore, increases in IMAT could create an inhospitable local environment that perpetuates the degradation of contractile properties that lead to losses in muscle function. A study of whether IMAT is related to the loss in muscle function will help to further characterize potential deleterious effects of changes in regional adipose tissue (7).

The use of medical imaging has greatly improved our ability to characterize human morphology for health and disease. Unfortunately, the emphasis has been placed on the characterization of muscle mass after reductions in activity, with little data to suggest a potential change in IMAT. A change in IMAT as a result of reduced activity may provide preliminary evidence that links sedentary behavior with an increased risk of metabolic disorders because higher levels of IMAT are associated with impaired glucose regulation. However, the change in IMAT may simply be explained by the loss in muscle. Nevertheless, the characterization of IMAT after a perturbation will spearhead efforts to characterize changes in a greatly understudied depot of adipose tissue.

We adopted an experimental model in which a single limb receives reduced activity while allowing participants to maintain mobility. We aimed to evaluate changes in IMAT of the thigh and calf after 4 wk of reduced activity and to determine whether these adaptations were related to changes in muscle strength.

SUBJECTS AND METHODS

Subjects and experimental design

Eighteen (6 men and 12 women) subjects completed the experimental protocol. Participants were recruited through flyers and presentations in the local University community and were paid for their efforts. Volunteers were excluded if they had a family history of blood clotting or smoked cigarettes.

Most studies of unilateral limb reduced activity have compared the immobile limb with the mobile limb (22). We did not use this design because the mobile limb is unlikely to be constant. Instead, participants underwent testing after a 4-wk control period and then again after the 4-wk reduced activity period. During each visit, magnetic resonance imaging (MRI) was undertaken and strength was assessed in the calf and thigh muscle groups. The Syracuse University and SUNY (Syracuse University New York) Upstate Medical University Institutional Review Boards approved all protocols, and the subjects provided written informed consent before participation.

Reduced activity model and compliance

We chose the unilateral limb suspension model because it allows subjects to maintain mobility while severely limiting activity in a single limb. This model was described previously (22). It requires subjects to use crutches while wearing a shoe with an elevated sole (10 cm) on the right foot, thus eliminating ground contact by the left foot.

We decreased the risk of venous thrombosis (23) by having subjects wear graduated compression stockings (24), administering aspirin (25), and asking them to elevate their left leg when possible. We also asked subjects to avoid air travel, sitting for long periods of time, crossing their legs, and drinking alcoholic beverages. We periodically examined the subjects for signs and symptoms of venous thrombosis (ie, redness, tenderness, localized warmth, and pitting edema).

Subject compliance was monitored with accelerometers, which were worn on the unloaded ankle. The accelerometers used (AMP-331; Dynastream Industries Inc, Alberta, Canada) inertial sensors that track motion in real time and uses this data to detect steps. We previously conducted experiments to validate this method for compliance during the protocol (26). The sensitivity of the accelerometer in detecting walking steps is 96.1%, and its specificity for not detecting steps during crutch ambulation is 96.5%. The participants wore the accelerometer for 3 d during the control period and then continuously during the entire 4-wk protocol. The subjects were provided transportation to classes to avoid injury while walking on crutches in the winter.

Soft tissue analysis

The participants were placed in a 1.5 Tesla scanner (Philips Medical Systems, Bothell, WA) where T1-weighted images were collected from the thigh and calf regions. Ten and 5 contiguous axial slices (10-mm thickness) were obtained at the midthigh and midcalf, respectively. Muscle, subcutaneous adipose tissue, and IMAT were measured volumetrically.

MIPAV (version 1.3; Medical Image Processing, Analysis and Visualization, Center for Information Technology, National Institutes of Health, Bethesda, MD) was used to analyze images on a personal computer workstation (27). IMAT was defined as the visible high-signal intensity (light) pixels between muscle groups and within muscle fascia. We used a modified version of 2 previously described strategies for measuring adipose tissue (28, 29). We first used a well-established nonparametric nonuniform intensity normalization (N3) algorithm that corrects smoothly varying shading caused by poor radiofrequency coil uniformity or gradient-driven eddy currents (30). This step is essential for subsequent analyses that assume images are homogeneous. Next, bone was removed and an investigator drew a rectangular region of interest containing $\approx$50% muscle and $\approx$50% subcutaneous adipose tissue. The rectangles were placed at 5 different areas around the thigh (or calf) producing 5 bimodal distributions of muscle and subcutaneous fat signal intensity peaks. The intensity value immediately to the right of the muscle histogram was chosen as the signal intensity threshold. The 5 threshold values were averaged and applied to all slices. All images were read in random order, and one investigator (MAN) performed the analyses. The technical error of identifying the signal intensity threshold, which allows separation of fat from muscle, is CV = 3.4% (n = 10), and the error associated with drawing the fascia latta border, separating IMAT and subcutaneous adipose tissue, is CV = 0.26% (n = 10).

Muscle strength

Knee extension and plantar flexor muscle strength were evaluated during maximal voluntary isometric contractions and used as an estimate of thigh and calf strength, respectively. The participants were first seated in a knee extension dynamometer (MedX, Ocala, FL) with their hip joint at 100 ° from flexion and a belt placed around the hip to prevent movement during contraction. The left leg knee joint was placed at 60 ° and secured with straps around the calf while force was measured at the axis

TABLE 1
Participant characteristics[1]

	Age	Weight	BMI	Number of steps	Total strength[2]	Total IMAT volume[3]	Total muscle volume[4]
	y	*kg*	*kg/m²*		*N*	*cm²*	*cm³*
Men ($n = 6$)	21.0 ± 3.5	66.2 ± 11	21.0 ± 3.8[5]	4321 ± 1107	982 ± 201[5]	160 ± 52.2[5]	1632 ± 332[5]
Women ($n = 12$)	20.8 ± 2.5	65.3 ± 10.9	25.2 ± 3.8	5273 ± 2135	710 ± 98	213 ± 45.3	1382.8 ± 166
Total ($n = 18$)	20.9 ± 2.8	65.6 ± 10.6	23.8 ± 4.2	4956 ± 1877	801 ± 188	196 ± 52.9	1465 ± 254

[1] All values are $\bar{x}$ ± SD. IMAT, intermuscular adipose tissue.
[2] Thigh + calf strengths for the average preinactivity testing periods.
[3] Thigh + calf IMAT volume for the average of preinactivity testing periods.
[4] Thigh + calf muscle volume for the average preinactivity testing periods.
[5] Significantly different from the women, $P \leq 0.05$ (ANOVA).

of rotation. To determine plantar flexor strength, the participants were positioned in a custom-modified dynamometer (Parabody 826; LifeFitness, Schiller Park, IL) with the hip, knee, and angle joints secured at 90°. During both protocols, the subjects performed multiple trials (3, 4) with 1–2 min of rest between trials. The maximal force recorded in kilograms was used for data analysis. The reliability of the strength tests in our laboratory is as follows: CV = 4.2% and intraclass correlation coefficient = 0.97 (31).

Data analysis

The sample size for this study was powered ($\beta > 0.80$, $\alpha = 0.05$) by using data from a previous study by our group, in which muscle strength and volume were estimated to decrease 16% and 11%, respectively (22). A 2-factor repeated-measures analysis of variance (ANOVA) (between-subjects factor: muscle group; within-subjects factor: time) was used to assess adaptations in soft tissue volumes (muscle, IMAT, and subcutaneous adipose) and muscle strength after the experimental protocols. We first performed the analyses during the control period. As expected, no significant changes were observed during the control period; thus, an average from the 2 control periods (average before inactivity) was calculated and used in separate analyses to compare with the values obtained after inactivity (average before inactivity versus average after inactivity). Significant muscle group (thigh versus calf) × time interactions were followed with a priori one-way repeated-measures ANOVA. A Bonferroni correction was used to control for overall type I error when multiple comparison tests were performed across the control period and the inactivity period. In the original study, the subjects were placed into 3 groups: control, applied ischemia, and motor imagery groups (14, 32). However, in the current study, the groups were combined because no differences in the change in IMAT across the interventions (intervention × time interaction: $P = 0.92$) were found. Furthermore, there was no theoretical rationale for why the interventions would induce changes in IMAT.

We created additive multiple regression models to determine whether the change in IMAT was related to the change in strength. Several methods are available for assessing change in regression models, and we chose a hierarchical approach by adding baseline values to help correct for regression to the mean. We also added the change in muscle volume because it is likely to be associated with strength change. No interaction ($P = 0.703$) was observed between muscle groups in this analysis; thus, volumes for the thigh and calf were combined to create a sample size of 36 observations for the regression model.

In addition to traditional methods, regression analyses were estimated by using clustering techniques that adjust for the lack of independence across observations. This procedure adjusts the SEs, and thus the P values, but does not alter the β coefficients. The β coefficients (SEs), P values, semipartial r^2 values, and unique amount of variance explained by each independent variable are provided in tables. An α level of significance was set at 0.05, and STATA 8.2 (StataCorp, College Station, TX) was used for all data analysis.

RESULTS

Subject characteristics and compliance

Participant characteristics are listed in **Table 1**. Compared with the women, the men were stronger, had a greater muscle volume, had a lower IMAT volume, and had a lower body mass index. Both the men and women showed similar effects over time; thus, all analyses were combined across sex. The experimental protocols were largely successful at reducing the number of steps on the left leg (4956 ± 1850 to 15.3 ± 10.7 steps, ie, a 99% decrease). As a result of reductions in activity, the subjects lost an average of 0.84 kg total body weight (65.6 ± 10.9 to 64.7 ± 10.7 kg; $P = 0.001$).

Soft tissue changes after reduced activity

The participants showed no significant changes in any of the tissue compartments of the thigh or calf over the control period (**Figure 1**). Muscle groups responded to lower levels of activity differently over time (muscle group × time interactions); thus, they were analyzed separately.

Compartmental changes in thigh and calf tissue as a result of reduced activity are shown in Figure 1 and **Table 2**. Four weeks of reduced activity caused a 2.8% and 3.7% reduction in total volume of the calf and thigh, respectively. Additionally, there was a 4.4% and 4.9% reduction in the fascia latta volume of the calf and thigh, respectively. Muscle volume decreased 7.4% and 7.7% in the thigh and calf, respectively (muscle group interaction: $P = 0.002$). IMAT was visibly higher after reduced activity (**Figure 2**) and increased disproportionally across muscle groups (muscle group interaction: $P = 0.02$).

The relative accumulation in IMAT exceeded the relative loss in both the thigh and calf regions (**Figure 3**). We performed a regression model to determine how much IMAT accumulation

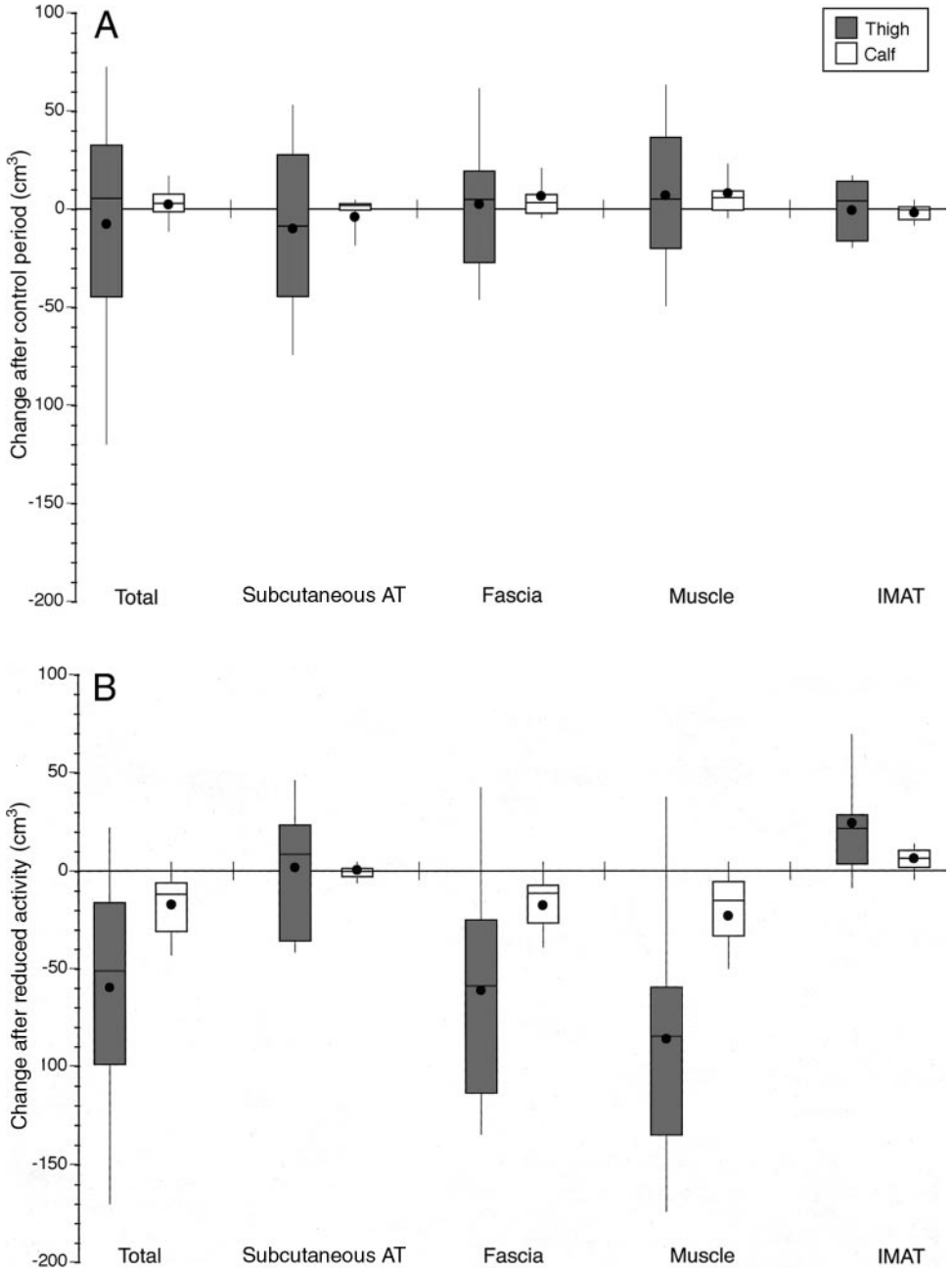

FIGURE 1. Box plots of absolute volume changes during a 4-wk control period compared with changes after 4 wk of reduced physical activity, ie, after unilateral lower limb suspension. $n = 18$. The top and bottom lines and the line through the middle of the box represent the 75th percentile (top quartile), 25th percentile (bottom quartile), and 50th percentile (median), respectively. The whiskers on the bottom extend from the 10th percentile (bottom decile) and top 90th percentile (top decile). The shaded circle represents the mean. AT, adipose tissue; IMAT, intermuscular AT.

may be explained by muscle loss. No interaction was observed between muscle groups; thus, the relation was examined with the muscle groups combined ($P = 0.481$). The loss in muscle explained 26% of the variance in IMAT accumulation after reduced activity (**Figure 4**).

Muscle strength

Thigh and calf strength did not change over the control period (thigh: 537 ± 33.8 to 547 ± 28.9 N, $P = 0.446$; calf: 248 ± 17.2 to 252 ± 20.0 N, $P = 0.453$). Reduced activity caused a 20% decrease in thigh strength and a 12.9% decrease in calf strength (muscle group × time interaction: $P < 0.001$; Table 2). With the thigh and calf volumes combined, regression analyses showed that the gain in IMAT was related to strength loss in model 1 (**Table 3**). Correction for baseline muscle strength attenuated the estimates (model 2), but adjustment for muscle volumes (change and baseline volumes) increased the statistical significance of IMAT (model 3). Overall, use of the semipartial r^2 values showed that the change in IMAT explained ≈4–6% of the change in muscle strength.

TABLE 2
Compartment volumes and muscle strength averaged across control periods and after reduced activity[1]

	Thigh			Calf		
	Average before inactivity	After inactivity	*P* (time effect)[2]	Average before inactivity	After inactivity	*P* (time effect)[2]
Total volume (cm^3)	2136 ± 481	2076 ± 474	0.003	474 ± 77.9	457 ± 85.3	0.001
Fascia latta volume (cm^3)	1383 ± 215	1322 ± 227	0.001	363 ± 62.4	346 ± 71.0	0.001
Subcutaneous fat volume (cm^3)	752 ± 449	754 ± 458	0.85	110 ± 56.6	111 ± 58.7	0.85
Muscle volume (cm^3)	1164 ± 205	1078 ± 216	0.001	301 ± 58	278 ± 67	0.001
Intermuscular adipose tissue volume (cm^3)	166 ± 50	190 ± 60	0.005	30 ± 6.9	36 ± 8.6	0.001
Strength (N)	553 ± 130	440 ± 100	0.001	247 ± 74.7	215 ± 68.3	0.001

[1] All values are $\bar{x}$ ± SD; $n = 18$.
[2] ANOVA.

DISCUSSION

This study investigated whether a localized reduction in physical activity results in IMAT accumulation and whether this change is related to changes in muscle strength. We showed that 4 wk of reduced activity caused an increase in IMAT that exceeded the relative loss in muscle and that IMAT accumulation differed across muscle groups. We also found that the gain in relative IMAT was related to loss in muscle strength.

Muscle loss and IMAT accumulation

Participants in the study lost an average of 1.2% of their body mass, but this was not associated with the loss in lean mass ($r = 0.005$) or gain in IMAT ($r = -0.05$). These data suggest that participants were in negative energy balance, and our findings our consistent with those of other studies that were not able to fully explain the loss in body mass after bed rest (33).

Research on the morphologic changes that occur with reductions in activity has typically focused on muscle mass, and we are unaware of studies that specifically investigated changes in IMAT. Two studies showed increased levels of subcutaneous adipose tissue after 30 d of immobilization (plaster casting) and 42 d of bed rest in healthy males (34, 35). Another study in a rabbit model showed an increase in IMAT of ≈6% after 6 wk of severing the supraspinatus muscle (35). Interestingly, the adipose tissue specifically accumulated between muscle fiber bundles and not within the muscle fibers themselves. Overall, the accumulation of IMAT has not been clearly studied after reduced activity, which has prevented direct comparisons with the literature.

Several parallel models of reduced activity investigated the accumulation of IMAT. For example, a 3-fold increase in thigh IMAT was observed after 8 y of a spinal cord injury when compared with weight matched healthy control subjects (10). In a posthemiparetic stroke model (>3 y), a 3% increase in low-density lean tissue in the thigh was observed; a 5% increase was observed when expressed relative to muscle mass. Also, stroke patients had no change in subcutaneous fat, but had a larger relative reduction in lean mass (7%) when compared with the accumulation of IMAT. Our study adds to these parallel models, without pathologic interference, and suggests that short-term reduced activity in healthy young men and women causes substantial (15–20%) increases in IMAT without changes in subcutaneous adipose tissue.

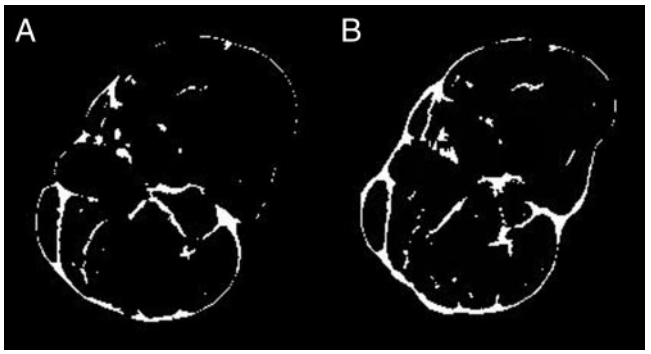

FIGURE 2. Illustration of the intermuscular adipose tissue of the thigh (middle slice of a 10-slice volume) before (A) and after (B) 4 wk of reduced activity, ie, after unilateral lower limb suspension. Intermuscular adipose tissue volume = 9.387 cm^2 (A) and 12.63 cm^2 (B). The white pixels represent intermuscular adipose tissue, and the black pixels represent background muscle and bone. Note that subcutaneous adipose tissue is not shown.

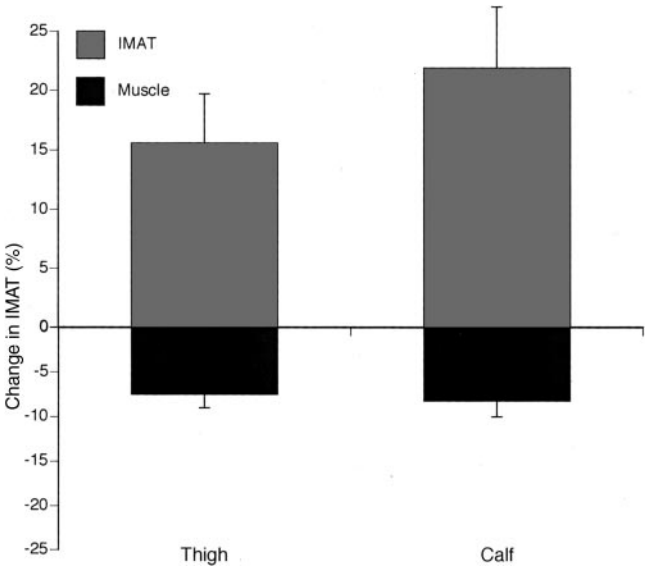

FIGURE 3. Percentage change in intermuscular adipose tissue (IMAT) and muscle in the calf and thigh regions after 4 wk of reduced physical activity, ie, after unilateral lower limb suspension. IMAT accumulation exceeded the loss in muscle and was greater in the calf than in the thigh. Results derived from ANOVA: muscle group × time interaction: $P = 0.02$. $n = 18$.

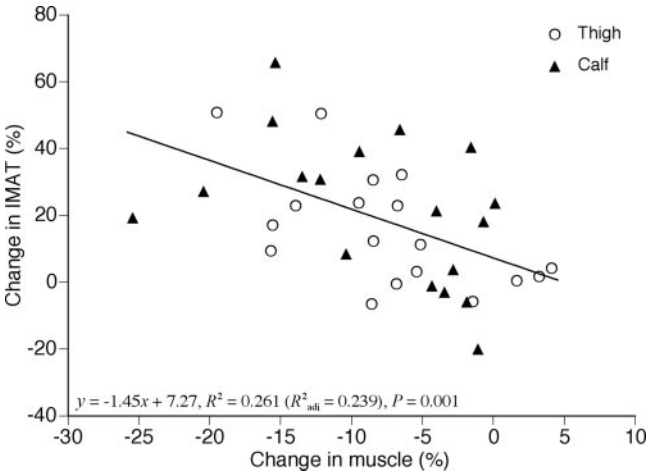

FIGURE 4. Percentage change in intermuscular adipose tissue (IMAT) as a function of the percentage change in muscle after 4 wk of reduced activity, ie, after unilateral lower limb suspension. $n = 18$. Muscle loss explained 26% ($P = 0.001$) of the variance in IMAT accumulation. Results were derived from linear regression analysis. The adjusted R^2 (R^2_{adj}) value accounts for sample size and number of independent variables according to the following equation: $R^2 - (k - 1)/(n - k) \times (1 - R^2)$, where n is the number of observations and k is the number of independent variables.

Physiologic explanations for increased IMAT

The accumulation of IMAT after a reduction in activity arises from either an excess triacylglycerol influx from the vasculature or altered fat oxidation in the muscle. Recent evidence suggests that physical inactivity blocks the uptake of plasma triacylglycerols by down-regulating lipoprotein lipase activity, which directly implicates altered fat oxidation as being responsible for inactivity-induced IMAT accumulation (3, 36).

The larger increase in IMAT relative to muscle loss supports a shift in fuel metabolism away from lipid toward glucose utilization commonly observed after inactivity (4). This shift in fuel metabolism is likely due to impaired mobilization of intramuscular triacylglycerols because reductions in concentrations of 3-hydroxyacyl CoA dehydrogenase, a key enzyme in fatty acid oxidation, are seen following bed rest (37, 38). Furthermore, denervation causes an increase in malonyl-CoA, which in turn inhibits caritine palmitoyl transferase I (CPT-I), a rate-limiting step for the transport of fatty acyl-CoA into the mitochondria (39). Impaired oxidative capacity though CPT-I and 3-hyroxyacyl CoA dehydrogenase coupled with reduced mitochondrial volume (38) likely actuates the transformation from oxidative to glycolytic muscle fibers during inactivity. The higher level of glycolytic muscle metabolism may cause a dysregulation of intramyocellular lipids that creates an environment to promote the visible accumulation of IMAT (4).

Interestingly, we found a greater relative accumulation of IMAT in the calf than in the thigh. This finding supports Hikida et al, who showed a greater reduction in oxidative enzyme activity in the soleus than in the vastus lateralis (37). Therefore, muscles with an initially higher oxidative capacity may have a greater propensity to accumulate IMAT.

Consequences of IMAT accumulation

There are no clear advantages to the accumulation of adipose tissue in atrophied muscles. In fact, clinically speaking, there seem to be several consequences, because higher concentrations of muscle lipid are linked to insulin resistance (6, 40, 41), an increased risk of physical limitation (12), and reduced strength in older adults (5). Thus, findings from this study raise a number of important issues regarding the etiology of IMAT.

A novel finding from this study was that strength loss was related to increases in IMAT even after correction for the change in muscle. Although our finding is certainly preliminary, it provides another potential consequence of IMAT accumulation and source for study. This association may be secondary to an effect on muscle function through adipose tissue's role as an endocrine organ (42). For example, TNF-α, a common cytokine expressed by adipose tissue (16–18), impairs force production independent of muscle wasting (19, 21, 43–45). This was first believed to be due to decreases in calcium concentrations from the sarcoplasmic reticulum (46) but is now thought to occur downstream of the calcium signal at the myofilament level (19). In support of this finding, our previous work suggests several adaptations in chemical signal transduction pathways and mechanical properties of inactive human muscle that affect strength loss (14, 32). All in all, an increase in IMAT could provide an inhospitable environment to promote contractile dysfunction, but further work is needed to verify this hypothesis

TABLE 3
Associations between changes in intermuscular adipose tissue (IMAT) and muscle strength from a linear regression analysis with thigh and calf combined[1]

	B (SE)	P	B (SE)[2]	P^2	Semipartial r^2	Full model R^2	Adjusted[3] R^2
Model 1						0.178	0.154
Change in IMAT volume (cm^3)	−1.20 (0.44)	0.010	−1.20 (0.28)	<0.001	0.178		
Model 2						0.611	0.588
Change in IMAT volume (cm^3)	−0.616 (0.32)	0.066	−0.616 (0.26)	0.029	0.043		
Baseline strength (N)	−0.256 (0.04)	<0.001	−0.256 (0.05)	<0.001	0.527		
Model 3						0.633	0.571
Change in IMAT volume (cm^3)	−0.960 (0.44)	0.039	−0.960 (0.49)	0.069	0.057		
Baseline strength (N)	−0.316 (0.09)	0.001	−0.316 (0.07)	<0.001	0.152		
Baseline IMAT volume (cm^3)	−0.031 (0.18)	0.865	−0.031 (0.22)	0.886	<0.001		
Change in muscle volume (cm^3)	−0.157 (0.17)	0.379	−0.157 (0.24)	0.528	0.010		
Baseline muscle volume (cm^3)	0.029 (0.05)	0.557	0.029 (0.07)	0.674	0.004		

[1] $n = 36$. IMAT, intermuscular adipose tissue; B, regression coefficient.
[2] Values adjusted for the lack of independence across muscle regions.
[3] Accounts for sample size and number of independent variables according to the following equation: $R^2 - (k-1)/(n-k) \times (1-R^2)$, where n is the number of observations and k is the number of independent variables.

MRI measurement of IMAT

Contrast differences in MR images are dictated by the density of hydrogen nuclei and relaxation times of the tissue. These differences in contrast were used to segment adipose from muscle tissue (or high- from low-signal intensity pixels). This method suffers from partial volume effects as a result of difficulties in determining when adipose exactly ends and muscle exactly begins. To prevent erroneous decisions, we standardized the image segmentation by having one investigator segment all images in a random order and followed previously established imaging segmentation protocols (28, 29). Furthermore, our cutoff signal intensity to segment muscle and adipose tissue did not change after the protocol ($P = 0.593$ and 0.677 for the thigh and calf, respectively). In further support, quantification of IMAT with MRI methods similar to this study is highly correlated with cadaver dissection ($r = 0.92$) (47).

Water shifts after reduced activity (33) could increase longitudinal MR relaxation (T1) times and thus increase pixel signal intensity. However, it is unlikely that T1 relaxation times changed in our study in light of stabilization in transverse relaxation times (T2) during fluid shifts (48, 49).

Study limitations

An obvious limitation of this study was that we used knee extension and calf flexion strength values to represent the strength of the entire volume of the respective soft tissue regions. We felt that this was appropriate considering that strength across muscle groups is highly correlated. The lack of muscle biopsy and blood samples was also a limitation of this study. Muscle biopsy samples would have helped us determine whether the muscles had higher concentrations of cytokines, as found with atrophied muscle after stroke (50), and if this was related to an accumulation of IMAT. Blood samples could also help determine whether the accumulation of IMAT was associated with changes in glucose disposal. Future work will include these measures.

Conclusions

IMAT shows marked increases after a short period of reduced activity in healthy young adults. Although the reduction in activity in this study was extreme, it provides preliminary evidence that implicates physical activity levels in the etiology of IMAT. Although several others have investigated IMAT in diabetic, lipodystrophic, HIV-infected, stroke, and spinal cord–injured patients, our study adds to the existing literature that suggests a role for reduced physical activity induced accumulation of IMAT in nonpathologic models.

TMM was responsible for the study concept, analyzed and interpreted the data, and wrote the manuscript. BCC conducted all aspects of the experimental protocols, provided the data from previous work (14, 32), provided intellectual support, interpreted the data, and critically reviewed the manuscript. MAN analyzed the MR images and critically reviewed the manuscript. BHG and LLP-S critically reviewed the manuscript and provided intellectual support. TBH contributed to the study concept, critically reviewed the manuscript, and provided intellectual support. Each author declared that he or she had no conflict of interest (financial or personal) in any company or organization sponsoring this study.

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