Sex-specific differences in leg fat uptake are revealed with a high-fat meal

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Votruba, Susanne B., and Michael D. Jensen. Sex-specific differences in leg fat uptake are revealed with a high-fat meal. Am J Physiol Endocrinol Metab 291: E1115–E1123, 2006.—The mechanism(s) by which sex specific differences in regional body fat distribution develop are not known. We assessed the effects of a high-fat (HF) meal on fatty acid oxidation and uptake into regional fat depots using isotopic tracers and adipose biopsies. Thirty men (BMI 23.6 ± 0.3 kg/m²) and 29 women (BMI 22.4 ± 0.3 kg/m²) received a meal containing [1H]triolein. Twelve of the men and 13 of the women received an additional 80 g of triolein in the meal (HF) and the remainder received a normal-fat (NF) meal. Adipose tissue lipoprotein lipase (LPL) activity was measured in the fed and fasted state. After 24 h, meal fatty acid uptake into subcutaneous adipose tissue was assessed. The efficiency of meal fat uptake into upper body subcutaneous fat was similar in both sexes, but women had a greater leg fat uptake, especially in response to a HF meal (P < 0.0001). A correlation between fed-state LPL activity and meal fat uptake was found in both upper and lower body fat (P < 0.0001, r = 0.69). These studies show that, in times of net fat storage, women preferentially increase uptake in leg adipose tissue, and this is likely mediated by fed-state LPL activity.

lipoprotein lipase; isotopic fat tracers; triolein; fat biopsies

HUMANS HAVE A REMARKABLE VARIABILITY in body fat distribution, and these differences have implications for metabolic health. For example, obese men and women with an upper body fat distribution are more likely to develop metabolic complications than those with a lower body fat distribution (22), including obesity-related dyslipidemia, hypertension, insulin resistance, and type 2 diabetes mellitus (21). Why some individuals preferentially store body fat in the central depots, whereas others store it in peripheral depots, is unknown, but there may be clues from the normal sexual dimorphism of fat distribution. With sexual maturity, men accumulate more fat in the upper body/visceral region and women accumulate more in the lower body region (36). The preferential accumulation of fat in some depots more so than others must be related to either regional differences in adipose tissue lipolysis and/or fatty acid uptake, but it is not yet clear which process(es) regulates fat distribution.

Our search for differences in regional lipolysis as a factor responsible for regional fat distribution (3, 15, 17, 18, 26) has not yielded evidence in favor of this hypothesis. We therefore have studied whether regional differences in meal fatty acid uptake may be a determinant of regional body fat distribution. We (20, 32, 41) and others (24, 25) have found that the upper body subcutaneous fat (UBSQ) depot takes up meal fat more efficiently than lower body subcutaneous fat (LBSQ) and that this is the case in both women and men (20, 25, 32, 41). Thus the regional differences in adipose tissue lipolysis and adipose meal fatty acid uptake do not give clues as to why net storage of body fat differs in these two regions between women and men.

The findings thus far suggest that upper body fat and lower body fat cells in men and women are managing triglyceride (TG) uptake and lipolysis in fundamentally similar ways. It is important to note, however, that all of our previous studies (32, 41) have been under conditions of isocaloric prestudy and study day diets. If regional variations in meal fatty acid uptake contribute to regional differences in body fat distribution, it may be that the differences become detectable only when net fat storage occurs, such as when meal fat intake exceeds the capacity for fat oxidation. This study was designed to assess regional meal fatty acid uptake in men and women under conditions of excess dietary fat to challenge the adipose storage system and mimic natural variations in fat intake that occur from day to day. We hypothesized that, compared with a normal meal, a greater proportion of the high-fat meal would be stored in body fat due to the limited ability of humans to increase fat oxidation as fat intake increases (9). We further hypothesized that the increased fat storage with the high-fat (HF) meal would preferentially be in the subcutaneous fat depots in women compared with men.

Lipoprotein lipase (LPL) is considered to be one of the key regulators of TG fatty acid uptake in adipose (4), and regional differences in LPL activity have been correlated with differences in fat distribution (30). Unfortunately, neither Márin et al. (23) nor our group (32) detected an association between regional LPL activity and regional meal fatty acid uptake despite early reports (13) that chylomicron uptake in rat adipose tissue was well correlated with LPL. However, both we (33) and others (7) have noted that the peak entry, and therefore peak exit, of labeled meal fat into and/or from the circulation occurs within 1 h after the ingestion of a second meal. This timing is important because adipose tissue endothelial LPL activity increases after meal ingestion (5, 42), and the activity at the time of peak meal fat disposal could be a better index of whether LPL is a key determinant of dietary fatty acid uptake (8). In this study, we performed adipose tissue biopsies both at the time of peak meal fatty acid entry into the circulation (for LPL activity) and 24 h after the ingestion of the isotopically labeled meal to measure fat storage. The results provide evidence that the female fat distribution pattern appears to be determined at times of net dietary fat storage and is strongly influenced by regional differences in meal-stimulated LPL activity.

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METHODS

Subjects. The Mayo Clinic Internal Review Board approved this study, and informed, written consent was obtained from a total of 30 lean men and 29 lean premenopausal women. No medications, with the exception of oral contraceptives, were allowed. Before study participation a complete blood count, chemistry group, and lipid profile were documented to be within normal limits. Participants were required to be weight stable for at least 2 mo before the start of the study.

Protocol. Body composition was assessed before the start of the study (see below). An outline of the study design is provided in Fig. 1. The volunteers were fed at the Mayo Clinic General Clinical Research Center (GCRC) metabolic kitchen for 3 days before the meal tracer study to ensure weight stability and constant macronutrient intake, as previously described (33, 41). Subjects were weighed each morning during the 3-day period, and energy intake was adjusted accordingly if weight changes were noted. Macronutrient content of the diet at this time was 50% carbohydrate, 35% fat, and 15% protein.

After the dietary control period the volunteers were admitted to the GCRC at 1700 and were given a research meal at 1800. Subjects were not allowed to eat again until the test meal the next morning and remained in the GCRC for two nights. Baseline blood and urine samples were obtained at 0745 the next morning, and at 0800 the participants were given the test meal. This liquid meal (Ensure Plus; Ross Laboratories) provided calories equaling either 40% of their resting energy expenditure (REE), as determined by indirect calorimetry [normal-fat (NF) meal] or 40% of REE plus an additional 80 g of triolein (HF meal; kindly provided by Karlshammar, Karlshammar, Sweden). The test meal contained ~80 μCi of [3H]triolein, which was sonicated into the Ensure Plus as previously described (32). For those receiving the HF meal we added the unlabeled triolein after the [3H]triolein had been sonicated into the Ensure Plus. Each participant received normal (solid food) meals at 1300 and 1800 based on their previous pattern of food intake. Blood samples were taken hourly until 1600 and then less frequently until 0800 the following morning. These samples were analyzed for plasma insulin and glucose concentrations as well as plasma chylomicron and nonchylomicron TG concentrations and specific activity (SA; see below). Adipose tissue biopsies (abdomen and thigh) for measurement of lipoprotein lipase activity were taken 1 h after lunch because that is the time of peak entry of the fatty acid tracer from breakfast into the bloodstream (32).

VO₂ and VCO₂ were measured with indirect calorimetry using the V̇max metabolic cart (Sensor Medics, Yorba Linda, CA) beginning just before the meal, hourly for 8 h after the meal, and then 10 h after the experimental meal. REE was measured the next morning before breakfast. Urine was quantitatively collected for 24 h to assess both 3H₂O losses and urinary nitrogen excretion. Twenty-four hours after the consumption of the test meal, contralateral needle biopsies of the abdominal (UBSQ) and femoral (LBSQ) subcutaneous adipose tissue were performed to assess [3H]triolein uptake into these depots and to measure LPL activity.

Materials. [9,10-3H]triolein was purchased from NEN Life Science Products (PerkinElmer, Boston, MA). 2H₂O was obtained from Iso-tech (Miamisburg, OH).

Assays and methods. Dual-energy X-ray absorptiometry (DEXA) (DPX-IQ; Lunar Radiation, Madison, WI) was used to determine total body fat, leg fat, fat-free mass (FFM), and leg FFM. A Beckman glucose analyzer (Beckman Instruments, Fullerton, CA) was used to measure plasma glucose concentrations, and plasma insulin concentrations were measured using a chemiluminescence method with the Access Ultrasensitive Immunoassay system (Beckman, Chaska, MN). Chylomicron TG concentrations were determined as previously described (32). Heparin releasable LPL activity was measured as described by Nilsson-Ehle and Schotz (27) and expressed as micromoles of FFA released per hour per gram of adipose tissue. Fat cell size was measured using photomicrographs as previously described (39).

The amount of meal [3H]triolein added to the meal was quantified by counting quadruplicate 50-μl meal samples on a scintillation counter. 3H SA was multiplied by meal volume, including the added, unlabeled triolein where appropriate, to determine total amount of tracer consumed (dpm/meal). The addition of 80 g of triolein to the Ensure Plus resulted in a heterogeneous mixture (oil and meal) that made it difficult to collect a representative sample of the meal to count for 3H. Because the procedures used to prepare the meals were identical up to the point of adding the unlabeled triolein, we used the average value of [3H]triolein added to the NF meals as the tracer content for each of the HF meals. The disadvantage of this approach is that, although the average group values will be correct for meal fatty acid disposition, the individual values will not be as accurate. Although the addition of the unlabeled triolein resulted in some oil droplets that could not be adequately mixed into the Ensure Plus, we ensured that the volunteers consumed the entire contents of the container and assumed that the meal fat, the unlabeled triolein, and the [3H]triolein tracer mixed equally during digestion and absorption. This assumption seems reasonable given the plasma chylomicron SA values for those receiving the HF and NF meals (see RESULTS).

Adipose tissue lipids were extracted using standard procedures (10) and adipose tissue TG specific activity was measured as previously described (33). Briefly, extracted lipids were weighed and counted on the scintillation counter to <2% counting error. The TG SA was calculated (dpm/g lipid) for each site. The percentage of tracer uptake, and therefore meal fat, into adipose tissue and the uptake of meal fatty acids into adipose tissue TG (mg of meal fat/g of adipose tissue lipid) were calculated as previously described (32).

The concentration of 3H₂O in urine water was assayed at baseline and at 24 h for the determination of cumulative meal fat oxidation (33). The total amount of tracer accounted for at 24 h was calculated as the sum of the UBSQ and LBSQ percentage of tracer uptake and the percentage of tracer oxidized. “Unaccounted for” tracer is the difference between 100% and the accounted for tracer.

Calculations and statistics. The total energy expenditure, fat, and carbohydrate oxidation over the first 10 h after the experimental meal, during which hourly indirect calorimetry measurements were performed, was calculated as previously described (11).

Fig. 1. Schematic of study protocol. GCRC, General Clinical Research Center. Subcutaneous adipose tissue biopsies were performed in the abdominal and femoral regions.
All data are presented as means ± SE and were analyzed using the JMP 5.1.1 statistical package (SAS Institute, Cary, NC). A P value of <0.05 was taken to be significant. The effects of meal fat content and sex on meal fat disposal were assessed for by analysis of variance (ANOVA). Plasma glucose, insulin, and chylomicron TG values were analyzed using repeated-measures ANOVA. In all cases, if significant differences were present, a nonpaired t-test was used for direct comparisons. Linear regression analysis was used to determine the relationship between regional LPL activity and regional meal fatty acid uptake as determined by the tracer.

RESULTS

Subject characteristics. The average age of all subjects combined was 31 ± 1 yr, and age did not differ significantly between groups. Anthropometric body composition and selected laboratory data are presented in Table 1. As anticipated, men were heavier, with more FFM and less body fat than women; the expected differences in regional fat mass were also present. The plasma concentrations of TGs and total and HDL cholesterol are also provided in Table 1. There were no significant differences for any of the above parameters between the HF vs. NF meal groups.

Plasma glucose and insulin concentrations did not differ significantly between groups (data not shown). The plasma chylomicron concentrations were higher in men than in women after both the low-fat and HF meal (Fig. 2, top). The chylomicron TG SA values are depicted in Fig. 2, bottom. Because the same amount of [3H]triolein was added to both the NF and HF meals, the meal and/or chylomicron TG SAs were reduced in the HF meal groups. The peak concentration of meal TG tracer in plasma occurred between 300 and 360 min after the breakfast meal (~1 h after the lunch meal).

Energy metabolism. The baseline respiratory exchange ratio (RER) on the morning of the experimental meal did not differ significantly between groups and was 0.84 ± 0.01, 0.83 ± 0.01, 0.84 ± 0.01, and 0.85 ± 0.01 for NF men, HF men, NF women, and HF women, respectively. Figure 3 depicts the time course of RER in each group. There was an effect of time and meal type (both P < 0.0005) on RER by repeated-measures ANOVA but no effect of sex. The HF meal groups had lower RER values and, therefore, greater whole body fat oxidation than the NF meal groups between 60 and 600 min postmeal.

Table 2 provides energy expenditure and substrate oxidation data for the first 10 h after the experimental meal. Energy expenditure was slightly greater after the HF meal in both men and women compared with the NF meal (P < 0.05). The HF meal resulted in major differences in fat and carbohydrate oxidation (Table 2). When adjusted for total energy expenditure, the percentage of energy coming from fat (29 ± 3 and 23 ± 3% for men and women, respectively) and carbohydrate (60 ± 3 and 63 ± 3% for men and women, respectively) did not differ between sexes. The percentage of energy expenditure from fat was greater after a HF (36 ± 3%) than a NF (19 ± 3%) meal (P < 0.0005). Average protein oxidation over 24 h did not differ by meal type or sex.

Meal fatty acid disposition. Cumulative oxidation of meal fatty acids as determined by generation of [3H]2O from [3H]triolein was 55 ± 3, 44 ± 4, 41 ± 3, and 27 ± 2% for NF men, HF men, NF women, and HF women, respectively (ANOVA P < 0.0001; Fig. 4); men oxidized a greater percentage of dietary fatty acids than women (P < 0.0001). The percentage of meal fat oxidized over 34 h was less (P < 0.0004) after the HF meal than the NF meal.

To account for the differences in fat content between HF and NF meals, the fractional tracer oxidation was multiplied by the total fat content of the experimental meal. The meal fat oxidized over 24 h was 9.4 ± 0.6, 42.1 ± 4.2, 5.7 ± 0.4, and 25.5 ± 2.1 g for NF men, HF men, NF women, and HF women, respectively (P < 0.0001). Men oxidized more meal fat than women (P < 0.0001), and the absolute amount of meal fat oxidized was greater with the HF meal (P < 0.0001 vs. NF meal). A significant interaction between sex and meal fat amount was also found (P < 0.005), indicating that the amount of meal fat oxidized after the HF meal vs. the NF meal was greater in men than in women.

Twenty-four hours after ingestion of the experimental meal, most dietary fat, as traced by the [3H]triolein, can be accounted for by oxidation or storage in subcutaneous fat, but a portion of the meal fat remains unaccounted for and is presumably stored in an unmeasured depot(s). In these studies, the sum of meal fatty acid oxidation plus uptake of meal fatty acids into subcutaneous adipose tissue averaged 75 ± 4, 60 ± 4, 87 ± 6, and 62 ± 4% for NF men, HF men, NF women, and HF women, respectively (P < 0.0005; Fig. 4). The percentage of dietary fat accounted for by oxidation and storage in subcutaneous fat was not statistically different in men and women, but with the HF meal a greater (P < 0.0001) proportion of dietary fat was unaccounted for after 24 h.

Regional meal fatty acid uptake. The efficiency of dietary fat storage into UBSQ and LBSQ (mg of meal fat/g adipose tissue lipid) for the different groups is provided in Table 3. The

Table 1. Subject characteristics

<table>
<thead>
<tr>
<th></th>
<th>Normal-Fat Meal</th>
<th>High-Fat Meal</th>
<th>P Value</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Females</td>
<td>Males</td>
<td></td>
<td>Sex</td>
<td>Meal</td>
</tr>
<tr>
<td>Age, yr</td>
<td>29.7±2.3</td>
<td>32.1±2.6</td>
<td></td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Weight, kg</td>
<td>62.1±1.7</td>
<td>78.7±2.1</td>
<td></td>
<td>&lt;0.0001</td>
<td></td>
</tr>
<tr>
<td>BMI</td>
<td>22.4±0.6</td>
<td>23.7±0.3</td>
<td>&lt;0.0001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>%Body fat</td>
<td>30.0±1.5</td>
<td>19.2±1.5</td>
<td>&lt;0.0001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>UBSQ, kg</td>
<td>9.7±0.8</td>
<td>7.1±1.0</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td></td>
</tr>
<tr>
<td>LBSQ, kg</td>
<td>7.5±0.5</td>
<td>4.8±0.4</td>
<td>&lt;0.0001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Visceral fat, kg</td>
<td>1.2±0.3</td>
<td>1.8±0.3</td>
<td>&lt;0.0001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HDL, mg/dl</td>
<td>60±4</td>
<td>42±2</td>
<td>&lt;0.0001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cholesterol, mg/dl</td>
<td>153±7</td>
<td>160±7</td>
<td>&lt;0.0005</td>
<td>&lt;0.0005</td>
<td></td>
</tr>
<tr>
<td>Triglycerides, mg/dl</td>
<td>74±6</td>
<td>88±5</td>
<td>&lt;0.0005</td>
<td>&lt;0.0005</td>
<td></td>
</tr>
</tbody>
</table>

Values are means ± SE. BMI, body mass index in kg/m²; UBSQ, upper body subcutaneous fat; LBSQ, lower body subcutaneous fat; NS, not significant.
amount of dietary fat stored in subcutaneous fat was significantly greater after the HF meal in both sexes, and meal fat uptake in UBSQ was similar in men and women. In contrast, dietary fat storage in LBSQ was significantly greater in women than men, largely due to greater incremental uptake in response to the HF meal in women. This was evidenced by a statistical interaction \( P < 0.01 \) between sex and meal type for meal fat storage in LBSQ. A similar trend was seen in the UBSQ adipose tissue \( P < 0.049 \). Fat cell size did not differ between groups by sex or by meal type (Table 3), and therefore, expression of meal fat uptake per fat cell was not calculated. LBSQ fat cell size was significantly larger than UBSQ fat cell size in all groups (at least \( P < 0.05 \)) except men fed the HF meal \( P = 0.14 \).

The percentage of meal fatty acids taken up in different adipose depots is depicted in Fig. 4. Overall ANOVA for meal fatty acid uptake in the UBSQ was significant \( P < 0.005 \). Women had a greater UBSQ uptake (24 ± 3 and 22 ± 2% for NF and HF, respectively) than men (13 ± 2 and 12 ± 3% for NF and HF, respectively, \( P < 0.0005 \)), but there was no effect of a HF meal. Meal fatty acid uptake into LBSQ was significantly different between groups by ANOVA \( P < 0.0001 \). Again, a significant effect \( P < 0.0001 \) of sex was noted, with women having more LBSQ meal fat uptake (22 ± 3 and 13 ± 2% for NF and HF, respectively) than men (7 ± 1 and 4 ± 1% for NF and HF, respectively). The percentage of meal fat that was taken up in the LBSQ was reduced \( P < 0.05 \) after a HF meal. Upper body subcutaneous adipose tissue took up a
greater percentage of the meal fat than LBSQ \( (P < 0.01 \text{ for females and } P < 0.0005 \text{ for males}).

If meal fatty acid oxidation is a primary driving process and adipose tissue uptake is essentially managing nonoxidized dietary fat, the differences we observed in fat oxidation could account for the differences in regional meal fatty acid uptake. We therefore recalculated the proportion of dietary fat stored in UBSQ and LBSQ using the nonoxidized fat as the denominator. Using this approach, uptake in the UBSQ region remained significantly greater in women (48 ± 5 and 25 ± 3\% for NF and HF, respectively) than in men (35 ± 5 and 16 ± 3\% for NF and HF, respectively; \( P < 0.05 \)) and less after a HF meal (\( P < 0.0001 \)). Likewise, LWSQ storage was significantly greater in women (38 ± 6 and 19 ± 3\% for NF and HF, respectively) than in men (16 ± 2 and 7 ± 1\% for NF and HF, respectively, \( P < 0.0001 \)); the percentage stored in LBSQ was reduced with the HF meal (\( P < 0.0005 \)).

Lipoprotein lipase activity. We had sufficient material to measure abdominal and/or femoral adipose tissue LPL activity in the fed state (afternoon biopsy) and in the fasted state (the 24-h biopsy) in 20 women and 20 men (Fig. 5). Fed LPL activity was greater \( (P < 0.0001) \) than fasted LPL activity in both men and women and in both abdomen and thigh. However, there were no significant differences in UBSQ LPL activity between groups. Femoral LPL activity was significantly greater in women than in men \( (P < 0.005 \text{ for fasted, } P < 0.005 \text{ for fed}) \), but there was no effect of meal type on LBSQ LPL activity. There was a strong correlation between UBSQ fed LPL activity and UBSQ meal fatty acid uptake \( (P < 0.0001, r = 0.65; \text{Fig. 6}) \) and between LBSQ fed LPL activity and LBSQ meal fat uptake \( (P < 0.0001, r = 0.70; \text{Fig. 6}) \). There was also a correlation between fasted and fed LPL activity for abdominal and femoral sites \( (r = 0.65, P < 0.0001) \).

**Discussion**

This study tested whether a high-fat meal, designed to perturb the balance between fat intake and oxidation, would uncover differences in regional fatty acid uptake between men

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**Table 2. Whole body substrate utilization**

<table>
<thead>
<tr>
<th>Substrate Use as %10-h EE</th>
<th>Normal-Fat Meal</th>
<th>High-Fat Meal</th>
<th>( P ) Values</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Females</td>
<td>Males</td>
<td>Females</td>
</tr>
<tr>
<td>Indirect calorimetry</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NPRQ, 10 h</td>
<td>0.94±0.01</td>
<td>0.92±0.01</td>
<td>0.89±0.01</td>
</tr>
<tr>
<td>REE, kcal/24 h</td>
<td>1,268±23</td>
<td>1,618±42</td>
<td>1,258±28</td>
</tr>
<tr>
<td>Test meal</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>kcal</td>
<td>433±10</td>
<td>540±14</td>
<td>1,273±12</td>
</tr>
<tr>
<td>Fat, g</td>
<td>13.7±0.3</td>
<td>17.1±0.4</td>
<td>97.5±0.4</td>
</tr>
<tr>
<td>10-h Substrate oxidation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Carbohydrate, g</td>
<td>108±6</td>
<td>132±7</td>
<td>90±5</td>
</tr>
<tr>
<td>Fat, g</td>
<td>10±2</td>
<td>18±3</td>
<td>22±3</td>
</tr>
<tr>
<td>Protein, g</td>
<td>20±2</td>
<td>22±3</td>
<td>18±2</td>
</tr>
<tr>
<td>Substrate use</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>use as %10-h EE</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Carbohydrate</td>
<td>72±3</td>
<td>68±4</td>
<td>57±4</td>
</tr>
<tr>
<td>Fat</td>
<td>15±3</td>
<td>21±3</td>
<td>31±4</td>
</tr>
<tr>
<td>Protein</td>
<td>14±1</td>
<td>11±1</td>
<td>11±1</td>
</tr>
</tbody>
</table>

Values are means ± SE. NPRQ, nonprotein respiratory quotient; REE, resting energy expenditure; EE, energy expenditure.
and women that have not been detected with standard meals. We hypothesized that, under conditions of dietary-induced net fat storage, women would preferentially increase subcutaneous meal fatty acid uptake and men would not. Although both men and women increased total fat oxidation and reduced proportions of meal fat stored in subcutaneous fat in response to a high-fat meal, there was an important difference in regional fat uptake. Compared with a normal-fat morning meal, the high-fat meal resulted in more efficient uptake of meal triglyceride in LBSQ adipose tissue in women than in men, whereas the uptake efficiency in UBSQ was not different in men and women. Thus, these studies show for the first time that, when meal fat consumption results in net fat storage, women preferentially increase uptake in LBSQ adipose tissue. The sex difference in postprandial adipose tissue LPL activity mirrored the sex differences in meal fatty acid storage. These findings have implications for sex-based differences in body fat distribution.

Past studies have shown that the men and women are nearly identical in terms of regional differences in lipolysis (3, 17, 18) and meal fatty acid uptake (32, 41) in vivo under energy and/or fat neutral conditions. In contrast, this study indicates that women store more fat in metabolically favorable depots when meal fat intake exceeds the body’s capacity to maintain a neutral fat balance. The observation that inter-individual differences in postprandial LPL activity are predictive of the proportionate meal fat stored in that depot strongly suggests that this enzyme is involved. We suggest that the postprandial translocation of intra-adipocyte LPL to the adipose tissue endothelium is a key step in determining regional fat storage and, potentially, body fat distribution.

The regulation of adipose tissue LPL activity is not completely understood. Our findings are consistent with the previously reported increase in adipose LPL activity after meal ingestion (5, 6, 42). The nutritional regulation of adipose tissue LPL activity seems to be under posttranscriptional and post-translational control (6, 35). Intracellular LPL can be degraded or modified to an active form (28) for transport and anchoring to the capillary endothelium, where it acts on circulating triglyceride-rich lipoproteins. The postprandial increases in insulin and glucose may combine to determine LPL activity (28, 35), but why leg and abdominal adipose tissue should respond differently in men and women is unknown. We found large inter-individual variations in the differences between post-absorptive vs. postprandial adipose tissue LPL activity, ranging from a 50% decrease to a 950% increase. The strong correlation between postprandial LPL activity and meal fat uptake suggests that these inter-individual differences play a significant role in determining regional fat storage. We found no previous studies comparing the regional regulation of LPL in response to nutrient intake in men vs. women or any attempts to link postprandial LPL activity to meal fatty acid uptake in humans.

Our results should be put in context with previous reports. For example, LPL has been reported to be either different (31) or the same (32) in men and women. In addition, regional LPL activity has been variably associated with regional differences in body fat distribution (30). The responsiveness in gluteal LPL

Table 3. Regional meal fat disposal

<table>
<thead>
<tr>
<th>Fat cell lipid, µg/cell</th>
<th>Normal-Fat Meal</th>
<th>High-Fat Meal</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Females</td>
<td>Males</td>
<td>Sex</td>
</tr>
<tr>
<td>UBSQ</td>
<td>0.38 ± 0.03</td>
<td>0.48 ± 0.05</td>
<td>0.1561</td>
</tr>
<tr>
<td>LBSQ</td>
<td>0.59 ± 0.05</td>
<td>0.59 ± 0.06</td>
<td>0.04395</td>
</tr>
<tr>
<td>Meal fat/g lipid, mg</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UBSQ</td>
<td>0.38 ± 0.05</td>
<td>0.46 ± 0.11</td>
<td>0.1288</td>
</tr>
<tr>
<td>LBSQ</td>
<td>0.38 ± 0.06</td>
<td>0.25 ± 0.03</td>
<td>0.04002</td>
</tr>
<tr>
<td>Meal fat/total fat mass, g</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UBSQ</td>
<td>3.5 ± 0.4</td>
<td>2.5 ± 0.4</td>
<td>0.0008</td>
</tr>
<tr>
<td>LBSQ</td>
<td>2.7 ± 0.4</td>
<td>1.1 ± 0.1</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

Values are means ± SE.
to meals has been reported to be affected by antecedent diet history, but there was no suggestion that differences between men and women were present (42). Regional meal fat uptake by isotopically labeled meal has not been found to be correlated with postabsorptive LPL activity (23, 32), whereas postprandial LPL was highly correlated with meal fatty acid uptake in this study. To the extent that postprandial LPL activity determines the ability of regional adipose tissue to take up circulating chylomicron triglycerides, the wide variability in responsiveness of adipose tissue LPL activity would explain the past difficulty in detecting such an association. Understanding what determines the sex-specific, region-specific, and interindividual differences in LPL activity responsiveness to nutrient intake may provide important clues as to how interindividual differences in regional fat gain evolve.

Consistent with previous studies (37), we found that a lesser fraction of meal fatty acids were oxidized after the high-fat meal than after the normal-fat meal; dietary fat oxidation was blunted by \( \sim 15\% \). Studies over longer time periods, however, have generally found that high-fat feeding increases fat oxidation within 1 wk (16, 40). We had anticipated that all added dietary fat would be shunted towards storage based on the report of Flatt et al. (9), who found that the addition of 50 g of butter fat to a meal did not change whole body fat oxidation over 9 h. Instead, we found that a single high-fat and high-energy meal substantially increases fat oxidation, a finding similar to that reported by Griffiths et al. (14). Both we and Griffiths et al. added 80 g of extra fat to the meal; we used triolein, whereas Griffiths et al. used mostly saturated fats. It may be that the lesser amount of added fat in the studies of Flatt et al. limited their ability to detect greater fat oxidation.

We found a substantial increase in unaccounted for meal fat tracer after the high-fat meal. In a previous study we could account for an average of 98\% of a meal fat uptake after 24 h if, in addition to measuring meal fat oxidation and uptake into subcutaneous adipose tissue, we determined uptake into visceral fat (20). We also found a correlation between unaccounted for meal fatty acids and amount of visceral fat (32), implying that the missing meal fatty acids may be going to the visceral fat depot. Because this depot is generally larger in men than in women (32), we hypothesized that only men would have a greater percentage of unaccounted for meal fat after the ingestion of a high-fat meal. Yet this phenomenon was seen in both sexes, although men fed the normal-fat meal had a higher percentage of unaccounted for tracer than women fed the normal-fat meal, again suggesting that at some level a sex difference in the trafficking of meal fat is present. We cannot be certain that the increase in unaccounted for meal fatty acids with the high-fat meal can be attributed to visceral fat uptake, however, because our previous studies (32, 41) have been only in normal-fat meals, and other sites of uptake (muscle and liver) cannot be excluded as significant sources of meal fat trafficking.

Data on the efficiency of triglyceride uptake into adipose tissue and adipose tissue LPL activity has been presented in a number of different ways. LPL activity has been expressed per gram of adipose tissue (1, 5, 30, 32, 38) as well as per fat cell (1, 2, 12, 30, 42). Expressing the efficiency of fatty acid uptake or LPL activity per fat cell provides information regarding cellular function per se but does not directly provide information regarding the tendency of depots to compete for triglyceride, especially if fat cell size differs between tissue beds. Conversely, although giving fatty acid uptake or LPL activity per gram of tissue allows comparison between tissue beds, it may not provide the context of adipocyte function. Here, we report data on meal fat uptake per gram of tissue but also give fat cell size characteristics of our volunteers, allowing readers to derive information most helpful to their specific needs. Of note, we confirmed the finding (2) that fasting LPL activity per cell generally increased as a function of fat cell size (data not shown), yet we found that fed LPL activity was less strongly related to fat cell size and meal fat uptake was largely unrelated to fat cell size.

In summary, these studies provide the first evidence that differences in regional meal fatty acid storage under conditions of net fat storage are associated with regional differences in fat
distribution in the expected direction in women vs. men. Furthermore, the strong correlation between postprandial adipose tissue LPL activity and regional meal fatty acid storage strongly suggests that this enzyme plays a key role in determining regional meal fat storage under conditions of excess fat intake. A better understanding of the posttranscripational and posttranslational regulation of adipocyte LPL handling, including environmental, hormonal, and genetic factors, should greatly improve our understanding of the determinants of body fat distribution in humans.

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