Excess body fat in men decreases plasma fatty acid availability and oxidation during endurance exercise

Bettina Mittendorfer, David A. Fields, and Samuel Klein

Mittendorfer, Bettina, David A. Fields, and Samuel Klein. Excess body fat in men decreases plasma fatty acid availability and oxidation during endurance exercise. Am J Physiol Endocrinol Metab 286: E354–E362, 2004. First published November 18, 2003; 10.1152/ajpendo.00301.2003.—The effect of relative body fat mass on exercise-induced stimulation of lipolysis and fatty acid oxidation was evaluated in 15 untrained men (5 lean, 5 overweight, and 5 obese with body mass indexes of 21 ± 1, 27 ± 1, and 34 ± 1 kg/m², respectively, and %body fat ranging from 12 to 32%). Palmitate and glycerol kinetics and substrate oxidation were assessed during 90 min of cycling at 50% peak aerobic capacity (V₀₂ peak) by use of stable isotope-labeled tracer infusion and indirect calorimetry. An inverse relationship was found between %body fat and exercise-induced increase in glycerol appearance rate relative to fat mass (r² = 0.74; P < 0.01). The increase in total fatty acid uptake during exercise [μmol/kg fat-free mass] × 90 min] was ~50% smaller in obese (181 ± 70; P < 0.05) and ~35% smaller in overweight (230 ± 71; P < 0.05) than in lean (354 ± 34 men). The percentage of total fatty acid oxidation derived from systemic plasma fatty acids decreased with increasing body fat, from 49 ± 3% in lean to 39 ± 4% in obese men (P < 0.05); conversely, the percentage of nonsystemic fatty acids, presumably derived from intramuscular and possibly plasma triglycerides, increased with increasing body fat (P < 0.05). We conclude that the lipolytic response to exercise decreases with increasing adiposity. The blunted increase in lipolytic rate in overweight and obese men compared with lean men limits the availability of plasma fatty acids as a fuel during exercise. However, the rate of total fat oxidation was similar in all groups because of a compensatory increase in the oxidation of nonsystemic fatty acids.

Obesity; lipolysis; substrate

ENDOGENOUS TRIGLYCERIDES (TG), stored in adipose tissue and muscle, are an important source of fuel for working muscles during exercise. Moderate-intensity exercise causes a two- to threefold increase in lipolysis of adipose tissue TG, which releases fatty acids into the bloodstream that are delivered to skeletal muscle (23, 31, 49, 53, 64). In addition, exercise stimulates lipolysis of intramuscular TG (4, 10, 25, 30), which release fatty acids that are directly oxidized by local mitochondria. The increase in lipolytic activity during exercise is primarily mediated by increased catecholamine release (1, 5, 20, 30, 32).

Obese persons have increased TG stores in adipose tissue and skeletal muscle (46), which contribute to many metabolic abnormalities associated with obesity (17, 46, 61). Therefore, endurance exercise, which stimulates the mobilization and oxidation of adipose tissue and skeletal muscle TG, may be a particularly useful approach for preventing and treating obesity-related metabolic diseases. However, the ability of obese persons to increase the use of endogenous TG during exercise may be limited, because they have a blunted catecholamine response to exercise (50) and decreased lipolytic sensitivity to catecholamine stimulation (6, 24).

Few studies have evaluated the effect of increased adiposity on lipid kinetics during exercise. The results from two studies demonstrated that the increase in palmitate rate of appearance (Rp) in plasma during moderate-intensity endurance exercise (45–55% peak aerobic capacity (V₀₂ peak)) is blunted in obese women compared with lean women (23, 27). However, the lipolytic response to exercise in overweight persons [body mass index (BMI) 25.0–29.9 kg/m²] or in obese men (BMI ≥30.0 kg/m²) has not been adequately studied. We recently found that there is sexual dimorphism in both resting lipolytic rates (37, 41) and the exercise-induced stimulation of lipolysis (38) in lean subjects. Therefore, it is possible that the lipolytic response to exercise in obese men may differ from that observed in obese women.

The purpose of the present study was to investigate the relationship between relative body fat mass and the exercise-induced stimulation of lipolysis and fatty acid oxidation. We hypothesized that the increases in the release of fatty acids into plasma and plasma fatty acid oxidation that occur during exercise are inversely related to percent body fat. Lipid kinetics were evaluated by using stable isotope-labeled tracer infusions in combination with indirect calorimetry in lean, overweight, and obese men at rest and during moderate-intensity endurance exercise.

METHODS

Subjects

A total of 15 untrained men (5 lean, 5 overweight, and 5 obese, defined by BMI values of 18.5–24.9, 25.0–29.9, and ≥30 kg/m², respectively), aged 25–45 yr (lean: 31 ± 3 yr; overweight: 37 ± 4 yr; obese: 38 ± 2 yr), participated in this study. Aerobic fitness, determined by V₀₂ peak, was not different between groups (Table 1). All subjects were considered to be in good health, except for obesity, after completing a comprehensive medical examination, which included a history and physical examination, a 12-lead electrocardiogram, and standard blood and urine tests. No subject was taking regular medications or smoked tobacco. All subjects had a stable body weight for ≥2 mo and had been sedentary (regular exercise <1 h/wk) for ≥6 mo before the study. Written informed consent was obtained from each subject before participation in the study, which was approved by the Human Studies Committee and the General Clinical Research Center (GCRC) Scientific Advisory Committee of Washington University School of Medicine in St. Louis, MO.

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
Characteristics of the study subjects

<table>
<thead>
<tr>
<th></th>
<th>Lean (n=10)</th>
<th>Overweight (n=10)</th>
<th>Obese (n=10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Height, cm</td>
<td>178±2 (171–182)</td>
<td>175±2 (168–180)</td>
<td>179±4 (169–190)</td>
</tr>
<tr>
<td>Weight, kg</td>
<td>67±4 (53–76)</td>
<td>85±4 (75–98)*</td>
<td>107±3 (96–113)*†</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>21±1 (18–23)</td>
<td>27±1 (26–29)*</td>
<td>34±1 (30–37)*†</td>
</tr>
<tr>
<td>Total fat-free mass, kg</td>
<td>55±3 (45–64)</td>
<td>63±3 (53–75)</td>
<td>74±3 (65–81)*†</td>
</tr>
<tr>
<td>Total fat mass, kg</td>
<td>11±2 (6–16)</td>
<td>22±1 (17–24)*</td>
<td>32±1 (29–35)*†</td>
</tr>
<tr>
<td>Total fat mass, % body wt</td>
<td>16±2 (12–21)</td>
<td>26±1 (22–29)*</td>
<td>30±1 (27–32)*†</td>
</tr>
<tr>
<td>Trunk fat mass, kg</td>
<td>5±2 (4–8)</td>
<td>10±1 (7–13)*</td>
<td>14±1 (12–17)*†</td>
</tr>
<tr>
<td>Trunk fat mass, %</td>
<td>16±3 (12–22)</td>
<td>25±2 (21–30)*</td>
<td>29±1 (26–31)*†</td>
</tr>
<tr>
<td>V̇O₂ peak, l/min</td>
<td>2.8±0.3 (2.1–3.5)</td>
<td>2.8±0.1 (2.6–3.3)</td>
<td>3.1±0.2 (2.3–3.7)</td>
</tr>
<tr>
<td>V̇O₂ peak, ml/kg FFM⁻¹min⁻¹</td>
<td>50±2 (46–55)</td>
<td>45±3 (36–53)</td>
<td>42±3 (34–52)</td>
</tr>
</tbody>
</table>

Values are means ± SE (range). V̇O₂ peak, peak aerobic capacity; FFM, fat-free mass. *Value significantly different from lean men, P < 0.05; †value significantly different from lean and overweight men, P < 0.05.

Experimental Protocol

Preliminary testing. Subjects were admitted to the outpatient unit of the GCRC in the evening, after they had fasted overnight. Total body fat mass (FM), fat-free mass (FFM), and truncal fat mass were determined by dual-energy X-ray absorptiometry, or DEXA (Hologic). Seven subjects were given a standard meal, containing 12 kcal/kg body wt of the GCRC in the evening before the isotope infusion study. At 0800 (~75 min), a priming dose of NaH¹³CO₃ (1.5 mol/kg) dissolved in 0.9% saline was given, immediately followed by a constant infusion of [¹³C]acetate (0.035 mol/kg⁻¹min⁻¹) dissolved in 0.9% saline; the labeled acetate infusion was maintained for 165 min (until the end of exercise). Breath samples were collected in evacuated tubes before the tracer infusion and at 60, 70, 80, and 90 min during exercise to determine ¹³CO₂ enrichment in expired breath, as previously described (23, 38).

Isotope infusion study. Subjects were admitted to the inpatient unit of the GCRC in the evening before the isotope infusion study. At 1900, they were given a standard meal, containing 12 kcal/kg body wt for lean subjects and 12 kcal/kg adjusted body wt (calculated as ideal body wt + 0.25 × (actual body wt − ideal body wt)) for overweight and obese subjects. The meal contained 55% of total energy carbohydrates, 30% fat, and 15% protein. At 2230, the subjects ingested a liquid snack (Ensure; Ross Laboratories, Columbus, OH), containing 80 g carbohydrates, 12.2 g fat, and 17.6 g protein, and then fasted until completion of the study on the following day. The following morning, at 0600, catheters were inserted into a forearm vein for isotope infusion and into a radial artery for blood sampling. At 0800 (~75 min), while the subjects were resting on a recumbent cycle ergometer (EC-C400R, Cateye, Osaka, Japan), priming doses of NaH¹³CO₃ (1.5 mol/kg) and [¹³C,²H]glycerol (1.5 mol/kg), dissolved in 0.9% saline, were administered, and constant infusions of [¹³C,²H]glycerol (0.1 mol·kg⁻¹·min⁻¹), dissolved in 0.9% saline, and [¹³C]palmitate (0.035 mol·kg⁻¹·min⁻¹), bound to human albumin (Centeon LLC, Kankakee, IL), were started and maintained for 165 min by use of calibrated syringe pumps (Harvard Apparatus, Natick, MA). All stable isotope-labeled tracers were purchased from Cambridge Isotope Laboratories (Andover, MA). From 0915 (0 min) until 1045 (90 min), subjects exercised at 50% of their V̇O₂ peak on the recumbent cycle ergometer. Recumbent cycling was chosen to enhance comfort and compliance.

Blood samples were obtained before the start of the isotope infusion, every 5 min from ~15 min to 0 min during resting conditions, and every 10 min during exercise (10–90 min) to determine glycerol and palmitate tracer-to-tracer ratios (TTRs) and plasma substrate and hormone concentrations. Blood samples were immediately transferred into: 1) chilled tubes containing EDTA to determine plasma fatty acid and glycerol concentrations and TTRs; 2) chilled tubes containing EDTA and aprotinin (Trasylol) to measure insulin concentration; and 3) chilled tubes containing reduced glutathione and EGTA to determine plasma catecholamine concentrations. Blood samples were placed in ice, and plasma was separated by centrifugation within 30 min of collection. Plasma was stored at −70°C until final analyses were performed.

Breath samples were collected in evacuated tubes before the tracer infusion and at 60, 70, 80, and 90 min during exercise to determine ¹³CO₂ enrichment in expired breath, as previously described (23, 38). Whole body oxygen consumption (V̇O₂) and carbon dioxide production (VCO₂) were determined at 0–15, 25–30, 55–63, 65–73, and 75–90 min during exercise by use of a metabolic cart (SensorMedics Vmax, Yorba Linda, CA), which was recalibrated between measurement periods.

Acetate infusion study. Six subjects (2 in each BMI group) completed an acetate infusion study, within 2 wk of the isotope infusion study, to determine the acetate correction factor needed to calculate plasma fatty acid oxidation rate (52, 55). Catheter insertion (into the forearm vein only) and the exercise protocol during the acetate infusion study were identical to the isotope infusion study protocol. At 0800 (~75 min), a priming dose of NaH¹³CO₃ (1.5 mol/kg) dissolved in 0.9% saline was given, immediately followed by a constant infusion of [¹³C]acetate (0.035 mol·kg⁻¹·min⁻¹) dissolved in 0.9% saline; the labeled acetate infusion was maintained for 165 min (until the end of exercise). Breath samples were collected in evacuated tubes before the tracer infusion and at 60, 70, 80, and 90 min during exercise to determine ¹³CO₂ enrichment in expired breath. Whole body V̇O₂ and VCO₂ were determined at 0–15, 25–30, 55–63, 65–73, and 75–90 min during exercise by use of a metabolic cart (SensorMedics Vmax), which was recalibrated between measurement periods.

Sample Analyses

Plasma insulin concentrations were measured by radioimmunoassay (19). Plasma catecholamine concentrations were determined by a single isotope-derivative radioenzymatic method (54). Plasma glycerol concentration was determined by gas chromatography-mass spectrometry (GC-MS) after addition of [²⁻¹³C]glycerol to plasma as an internal standard (23, 37). Plasma fatty acid concentrations were quantified by gas chromatography ( Hewlett-Packard 5890-II, Palo Alto, CA) after addition of heptadecanoic acid to plasma as an internal standard (44).

Plasma palmitate and glycerol TTRs were determined by GC-MS (MSD 5973 system with capillary column; Hewlett-Packard) as previously described (43, 44). Plasma proteins were precipitated with ice-cold acetone, and hexane was used to extract plasma lipids. Free fatty acids were converted to their methyl esters with iodomethane and isolated by use of solid-phase extraction columns. Ions at mass-to-charge ratio (m/z) 270.2 and 271.2, produced by electron impact (EI) ionization, were selectively monitored. The aqueous phase, containing glycerol, was dried by speed-vac centrifugation (Savant Instruments, Farmingdale, NY). Heptadecanoic acid (HFB) anhydride was used to form an HFB derivative of glycerol, and ions were produced by EI ionization. Glycerol concentration and TTR were determined by selectively monitoring ions at m/z 253, 254, and 257. The ¹³CO₂-to-¹²CO₂ ratio in expired air was determined by IRMS (Sira II, VG Fisons, Cheshire, UK) as previously described (23).

AJP-Endocrinol Metab • VOL 286 • MARCH 2004 • www.ajpendo.org
E356  LIPID KINETICS DURING EXERCISE

Table 2. Exercise workload and intensity in lean, overweight, and obese subjects

<table>
<thead>
<tr>
<th></th>
<th>Lean</th>
<th>Overweight</th>
<th>Obese</th>
</tr>
</thead>
<tbody>
<tr>
<td>Power output, W</td>
<td>79±13 (50–120)</td>
<td>75±9 (50–105)</td>
<td>73±11 (50–115)</td>
</tr>
<tr>
<td>Relative intensity, % VO2_peak</td>
<td>53±4 (45–55)</td>
<td>51±2 (45–55)</td>
<td>53±2 (47–56)</td>
</tr>
<tr>
<td>Oxygen consumption, ml/kg FFM 1/min</td>
<td>26±2 (21–30)</td>
<td>23±2 (19–29)</td>
<td>22±2 (19–28)</td>
</tr>
</tbody>
</table>

Values are means ± SE (range) during the last 30 min of exercise.

Calculations

**Palmitate and glycerol kinetics.** Palmitate Ra provides an index of plasma fatty acid availability; during catecholamine-stimulated lipolysis, palmitate Ra provides a measure of fatty acids released primarily from hydrolysis of adipose tissue TG (39). Palmitate rate of disappearance (Ra) provides an index of plasma fatty acid tissue uptake. Glycerol Ra provides an index of whole body lipolytic rate and measures the rate at which glycerol is released into the systemic circulation, presumably from hydrolysis of adipose tissue and intramuscular and plasma TG.

During resting conditions, palmitate and glycerol Ra in plasma was calculated by dividing the tracer infusion rate by the average arterial palmitate or glycerol TTR obtained between −15 and 0 min (Steele’s equation for steady-state conditions) (56). Palmitate Ra was assumed to be equal to palmitate Ra during rest. During exercise, glycerol Ra and palmitate Ra and Ra were calculated by using Steele’s equation for non-steady-state conditions (13, 56). The effective volume of distribution was assumed to be 55 ml/kg FFM for palmitate (26) and 300 ml/kg body wt for glycerol (2). However, we found that even a 50% error in the estimated effective volume of distribution would cause <5% change in calculated Ra and Rd because of the minimal changes in TTR between samples in our study.

The total lipolytic response to exercise was calculated as the area under the palmitate or glycerol Ra curve above baseline. The exercise-induced increase in tissue fatty acid uptake from plasma was calculated as the area under the palmitate Ra curve during exercise above baseline divided by the proportional contribution of palmitate to total plasma fatty acid concentration.

Substrate oxidation. Total fat and carbohydrate oxidation rates were calculated on the basis of Vo2 and VCO2 values obtained during the last 30 min of exercise, as previously described (12). Plasma palmitate oxidation rate was calculated by dividing the rate of appearance of 13C2O2 in expired breath (VCO2 × CO2 TTR) by plasma palmitate TTR. This value was corrected for the average 13C2O2 recovery (52, 55) determined during the acetate infusion study for each subgroup of subjects (lean, overweight, and obese). The rate of plasma fatty acid oxidation was calculated by dividing the rate of plasma palmitate oxidation by the proportional contribution of palmitate to total plasma fatty acid concentration. The oxidation rate of nonpalmitic fatty acids was calculated as the difference between the rates of total fatty acid oxidation, determined by indirect calorimetry, and plasma fatty acid oxidation as determined by isotope tracers. We assumed that intramuscular TG were the primary source of oxidized nonpalmitic fatty acids and that plasma TG were not an important source of fuel during exercise (29, 34, 62).

Statistical Analyses

A one-way analysis of variance (ANOVA) was used to test for differences in aerobic fitness, Vo2, and total fat oxidation at the end of exercise between groups. A two-way ANOVA (group × time) with repeated measures was performed to test the significance of differences in the exercise-induced changes in substrate kinetics and hormone concentrations between lean, overweight, and obese men. Significant F ratios from the ANOVA were followed by Tukey’s post hoc analyses. A Student’s t-test for independent samples was used to test the significance of differences in total lipid kinetics during exercise (area under the curve) between lean and overweight or obese men (overweight and obese groups were pooled because the exercise-induced response in these groups was not different). Pearson product-moment correlation was used to assess the relationship between the lipolytic response to exercise and adiposity (%body fat and BMI) and the proportional contribution of systemic plasma and nonsystemic fatty acids to total fatty acid oxidation. Student’s t-test for paired samples was used to test the significance of differences in total Ra and Rd during exercise and the contribution of plasma and nonplasma fatty acids to total fat oxidation within each group. A P value of =0.05 was considered to be statistically significant. All data are expressed as means ± SE.

RESULTS

Exercise Bout

Absolute power output, Vo2, and VCO2 were constant between 30 and 90 min of exercise (data not shown). Exercise workload, Vo2 relative to FFM, and relative intensity (%VO2_peak) were the same in all groups (Table 2).

Plasma Hormone Concentrations

Plasma insulin concentrations at rest and during exercise tended to be greater in overweight and obese than in lean men (Table 3), but the difference did not reach statistical significance (P = 0.09). However, the relative decrease in plasma insulin concentration from the beginning until the end of exercise was not different between groups. Plasma epinephrine concentration was not different between groups at rest. However, the increase in plasma epinephrine concentration during exercise was smaller in obese than in lean men; the total (overweight and obese groups were pooled because the exercise-induced response in these groups was not different). Pearson product-moment correlation was used to assess the relationship between the lipolytic response to exercise and adiposity (%body fat and BMI) and the proportional contribution of systemic plasma and nonsystemic fatty acids to total fatty acid oxidation. Student’s t-test for paired samples was used to test the significance of differences in total Ra and Rd during exercise and the contribution of plasma and nonplasma fatty acids to total fat oxidation within each group. A P value of =0.05 was considered to be statistically significant. All data are expressed as means ± SE.

Table 3. Plasma hormone concentrations during rest and exercise

<table>
<thead>
<tr>
<th></th>
<th>Lean</th>
<th>Overweight</th>
<th>Obese</th>
</tr>
</thead>
<tbody>
<tr>
<td>Insulin, μU/ml</td>
<td>5.7±0.7†</td>
<td>11.6±1.4†</td>
<td>20.8±7.5†</td>
</tr>
<tr>
<td>Epinephrine, pg/ml</td>
<td>56±12†</td>
<td>57±8</td>
<td>40±9</td>
</tr>
<tr>
<td>Norepinephrine, ng/ml</td>
<td>199±16†</td>
<td>264±28†</td>
<td>256±28†</td>
</tr>
</tbody>
</table>

Values are means ± SE. †Value significantly different from corresponding value at rest, P < 0.05; ‡value significantly different from corresponding value at rest, P < 0.05.
epinephrine response (area under the plasma epinephrine concentration curve above baseline, pg·ml$^{-1}$·90 min) decreased with increasing adiposity, from 3,129 ± 817 in lean to 2,004 ± 648 in overweight, and 1,116 ± 489 in obese men (obese vs. lean men, $P < 0.05$). Mean plasma norepinephrine concentrations at rest tended to be greater in overweight and obese than in lean men, but the difference did not reach statistical significance ($P = 0.10$); however, the relative increase in plasma norepinephrine concentration during exercise was similar in all groups.

Plasma Free Fatty Acid Concentrations

Total plasma free fatty acid concentration at rest was not different between lean (465 ± 83 μM), overweight (325 ± 34 μM), and obese (437 ± 31 μM) men. Plasma fatty acid concentrations decreased in all groups during the first 20–30 min of exercise and progressively increased thereafter, to 848 ± 54 μM in lean, 661 ± 120 μM in overweight, and 594 ± 78 μM in obese men at the end of exercise (obese vs. lean, $P = 0.055$). The contribution of palmitate to total plasma fatty acid concentration during resting conditions was the same in lean (27 ± 1%), overweight (27 ± 1%), and obese (28 ± 1%) men and did not change during exercise.

Palmitate and Glycerol Kinetics

During resting conditions, palmitate $R_d$ expressed per kilogram body weight or FFM was not different between groups. However, palmitate $R_a$ expressed per kilogram FM was greater in lean than in overweight ($P = 0.05$) or obese ($P < 0.05$) men (Table 4). Similarly, glycerol $R_a$ expressed per kilogram body weight or FFM was not different between groups, and glycerol $R_a$ expressed per kilogram FM was greater in lean than in obese men ($P < 0.05$) and tended to be greater in lean than in overweight men ($P = 0.059$; Table 4).

Exercise increased ($P < 0.01$ compared with resting values) palmitate $R_a$, (Fig. 1) and glycerol $R_a$ (to $45 \pm 7$, $23 \pm 3$, and $18 \pm 3$ μmol·kg FM$^{-1}$·min$^{-1}$ in lean, overweight, and obese men, respectively) in all subjects. However, the total increase (area under the curve above baseline) in palmitate $R_a$ ($92 \pm 10$, $50 \pm 15$, and $36 \pm 14$ μmol·kg·body wt$^{-1}$·90 min and $587 \pm 79$, $188 \pm 48$, and $119 \pm 43$ μmol·kg·FM$^{-1}$·90 min in lean, overweight, and obese men, respectively) and glycerol $R_a$ ($258 \pm 31$, $171 \pm 30$, and $175 \pm 40$ μmol·kg·body wt$^{-1}$·90 min and $1,656 \pm 232$, $667 \pm 101$, and $402 \pm 26$ μmol·kg·FM$^{-1}$·90 min in lean, overweight, and obese men, respectively) during 90 min of exercise was blunted in obese and overweight compared with lean men. Moreover, the increase in palmitate or glycerol $R_a$ during exercise was inversely correlated with percent body fat ($P < 0.01$) (Fig. 2) and BMI ($P < 0.01$; data not shown).

During resting conditions, palmitate $R_d$, expressed per kilogram body weight or FFM, was not different between groups. Exercise increased palmitate $R_d$ in all groups ($P < 0.01$), but the magnitude of the increase in palmitate $R_d$ above baseline was inversely related to adiposity (Fig. 3). In addition, the relationship between fatty acid $R_a$ and $R_d$ differed between lean and overweight or obese subjects. In lean men, total fatty acid $R_d$ during exercise was 15% lower than total fatty acid $R_a$ ($P < 0.05$); however, total fatty acid $R_d$ was not different from total fatty acid $R_a$ in overweight and obese men.

The relationship between plasma epinephrine concentration and palmitate $R_a$ is shown in Fig. 4. Maximal plasma epinephrine concentration during exercise decreased with increasing adiposity. In addition, at any given plasma epinephrine concentration during exercise (above resting values), palmitate $R_a$ was greater in lean subjects than in overweight or obese subjects.

Table 4. Palmitate and glycerol kinetics at rest

<table>
<thead>
<tr>
<th></th>
<th>Lean</th>
<th>Overweight</th>
<th>Obese</th>
</tr>
</thead>
<tbody>
<tr>
<td>Palmitate $R_a$</td>
<td>$\mu$mol·kg body wt$^{-1}$·min$^{-1}$</td>
<td>$1.0 \pm 0.1$</td>
<td>$0.7 \pm 0.1$</td>
</tr>
<tr>
<td></td>
<td>$\mu$mol·kg FFM$^{-1}$·min$^{-1}$</td>
<td>$1.2 \pm 0.2$</td>
<td>$1.0 \pm 0.2$</td>
</tr>
<tr>
<td></td>
<td>$\mu$mol·kg FM$^{-1}$·min$^{-1}$</td>
<td>$6.8 \pm 1.8$</td>
<td>$2.7 \pm 0.3^*$</td>
</tr>
<tr>
<td>Glycerol $R_a$</td>
<td>$\mu$mol·kg body wt$^{-1}$·min$^{-1}$</td>
<td>$2.1 \pm 0.4$</td>
<td>$1.9 \pm 0.4$</td>
</tr>
<tr>
<td></td>
<td>$\mu$mol·kg FFM$^{-1}$·min$^{-1}$</td>
<td>$2.5 \pm 0.5$</td>
<td>$2.6 \pm 0.6$</td>
</tr>
<tr>
<td></td>
<td>$\mu$mol·kg FM$^{-1}$·min$^{-1}$</td>
<td>$13.7 \pm 3.4$</td>
<td>$7.3 \pm 1.4^*$</td>
</tr>
</tbody>
</table>

Values are means ± SE. $R_a$, appearance rate; FFM, fat mass. *Value significantly different from lean value; $P < 0.05$; †tends to be different from lean value, $P = 0.059$. 

Fig. 1. Exercise-induced increase in palmitate appearance rate ($R_a$) relative to fat mass (FM, top) and total increase (relative with compared rest) during 90 min (bottom) in lean, overweight, and obese men ($n = 5$ per group). ANOVA revealed significant effects of time ($P < 0.001$), adiposity ($P < 0.001$), and adiposity × time interaction ($P < 0.001$) for the exercise-induced increase in palmitate $R_a$; post hoc analysis revealed no difference between overweight and obese subjects.

AJP-Endocrinol Metab • VOL 286 • MARCH 2004 • www.ajpendo.org
Substrate Oxidation During Exercise

The rate of total fat oxidation, assessed by indirect calorimetry during the last 30 min of exercise, was not significantly different between groups (lean: 6.1 ± 0.5; overweight: 4.9 ± 1.2; obese: 5.5 ± 0.5 mg·kg FFM⁻¹·min⁻¹). The oxidation of fat provided ~30% of total energy requirements during exercise in lean, overweight, and obese men. However, the source of fatty acids used as fuel during exercise varied between groups. In lean subjects, about one-half of the fatty acids oxidized during exercise were derived from systemic plasma fatty acids and the other one-half from nonsystemic fatty acids. The relative contribution of systemic plasma fatty acids to total fat oxidation decreased and the relative contribution of nonsystemic fatty acids, presumably derived primarily from lipolysis of intramuscular TG, increased with increasing adiposity. Therefore, the blunted lipolytic response to exercise in overweight and obese men compared with lean men limited the availability of plasma fatty acids for oxidation by working muscle during exercise. However, an increased oxidation of nonsystemic fatty acids compensated for the reduction in plasma fatty acid oxidation in overweight and obese men, so total fat oxidation during exercise was similar in all three groups.

DISCUSSION

Exercise is commonly recommended for overweight and obese persons to reduce body fat and improve metabolic health. In this study, we evaluated the effect of progressive increases in body fat on lipid metabolism during endurance exercise in sedentary men. Lipid kinetics were evaluated by using stable isotope-labeled tracer methods in conjunction with indirect calorimetry during rest and moderate-intensity cycling exercise in lean, overweight, and obese men, who were matched in age and aerobic fitness to eliminate the potential confounding influence of these factors on substrate metabolism. Our data demonstrate an inverse relationship between adiposity and the lipolytic response (increase in palmitate and glycerol Ra) to exercise. In addition, the oxidation of systemic plasma fatty acids, presumably derived primarily from lipolysis of adipose tissue TG, decreased, and the oxidation of nonsystemic fatty acids, presumably derived primarily from lipolysis of intramuscular TG, increased with increasing adiposity. Therefore, the blunted lipolytic response to exercise in overweight and obese men compared with lean men limited the availability of plasma fatty acids for oxidation by working muscle during exercise. However, an increased oxidation of nonsystemic fatty acids compensated for the reduction in plasma fatty acid oxidation in overweight and obese men, so total fat oxidation during exercise was similar in all three groups.
It is likely that differences in the increase in plasma epinephrine concentration during exercise between lean, overweight, and obese subjects contributed to differences in lipolytic rates between groups. Peak plasma epinephrine concentrations during exercise decreased as percent body fat increased. The mechanism responsible for the attenuated increase in plasma epinephrine concentration in our overweight and obese men is not known; however, this phenomenon has been observed previously in obese subjects by other investigators (50, 58). In our study, exercise intensity and degree of aerobic fitness, which are major determinants of the catecholamine response to exercise (7, 47), were similar in lean, overweight, and obese men.

Our data also suggest that an alteration in adipose tissue lipolytic sensitivity to circulating catecholamines contributed to the blunted lipolytic response to exercise in overweight and obese subjects. At any given plasma epinephrine concentration during exercise, palmitate Ra was much lower in overweight and obese subjects than in lean subjects. These results are consistent with data from previous studies that found the increase in fatty acid or glycerol Ra during epinephrine infusion blunted in obese compared with lean subjects (6, 24). The effect of plasma catecholamines on lipolysis is determined by the balance between the activation of \( \beta \)-adrenergic receptors, which stimulate lipolysis (1, 21, 40), and \( \alpha \)-adrenergic receptors, which inhibit lipolysis (21, 59). The potential mechanisms responsible for the blunted lipolytic response to catecholamines are controversial because of inconsistent results among studies. However, the data from most studies conducted in men suggest that increased \( \alpha \)-adrenergic receptor stimulation, rather than impaired sensitivity to \( \beta \)-adrenergic receptor stimulation, contributed to the reduced net lipolytic response to exercise in our overweight and obese subjects. Studies conducted in isolated subcutaneous abdominal adipocytes in vitro or in regional subcutaneous abdominal adipose tissue in situ found that lipolysis induced by \( \beta_1 \), \( \beta_2 \), and nonspecific \( \beta \)-adrenergic receptor stimulation was the same in lean and obese men (22, 35, 51). In contrast, the antilipolytic effect of \( \alpha_2 \)-adrenergic stimulation was greater in adipocytes isolated from obese than from lean men (35). Moreover, there is greater activation of adipose tissue \( \alpha_2 \)-adrenergic receptors during exercise in obese than in lean men (58). Data from weight loss studies support the notion that the alteration in lipolytic sensitivity in obese subjects is caused by excessive amounts of body fat, because diet-induced weight loss can correct the abnormality in catecholamine-stimulated lipolysis (36).

It is possible that “hyperinsulinemia” in our overweight and obese compared with our lean men contributed also to the blunted lipolytic response to exercise. Although the relative decrease in plasma insulin concentration was similar in all groups, the absolute plasma insulin concentrations were greater during exercise in overweight and obese than in lean subjects.

The difference in the lipolytic response to exercise between lean and obese men observed in our study is similar to findings.
made in obese women. We (23) and others (27) have found that the relative increase in fatty acid $R_a$ induced by endurance exercise is smaller in obese than in lean women. However, the mechanism responsible for the blunted lipolytic response to exercise associated with obesity may differ in men and women. In contrast to the findings from the present study and other studies that were conducted in men (50, 58), the increase in plasma epinephrine concentration during moderate-intensity exercise is the same in lean and obese women (11, 23, 27). Furthermore, unlike obese men (22, 35, 51), adipose tissue $\beta$-adrenergic receptor sensitivity is decreased in obese women (36, 48), whereas $\alpha_2$-adrenergic receptor stimulation during exercise is greater in both obese men and women compared with lean subjects (60). Therefore, the composite of these data suggests that there is sexual dimorphism in the adrenal medullary response to exercise and in adipose tissue lipolytic response to epinephrine stimulation in obese subjects. Nevertheless, the overall lipolytic response to exercise is blunted in obese subjects, both men and women.

Total exercise-induced increase in plasma fatty acid tissue uptake (fatty acid $R_a$ per kg FFM) was lower in our overweight and obese subjects than in our lean subjects, presumably because of the blunted increase in fatty acid release into plasma in the overweight and obese groups. Fatty acid availability was greater than fatty acid uptake in lean subjects, whereas availability and uptake were closely matched in overweight and obese subjects. Although whole body fatty acid uptake during prolonged moderate-intensity exercise depends on the availability of fatty acids from plasma (18, 40, 42, 62), muscle fatty acid uptake is a saturable, carrier-mediated process (3, 63). Therefore, our data suggest that the high rate of lipolysis during exercise in lean subjects may overwhelm cellular fatty acid transport systems, which leads to an increase in plasma fatty acid concentrations, whereas uptake of plasma fatty acids is not rate limiting in overweight or obese subjects because of their low rate of lipolysis.

Total fat oxidation during exercise was similar in our lean, overweight, and obese subjects. This finding is consistent with the results of some (14, 27, 57) but not all (16, 23, 28, 45) studies that investigated the effect of body fatness on fat oxidation during endurance exercise. The reason for the discrepancy among studies is not clear. In our study, we carefully controlled and matched the factors that influence fat oxidation during exercise across groups, including relative exercise intensity (49), total energy requirements (65), and aerobic fitness capacity (15).

Although the total fat oxidation rate was the same in all groups in our study, the source of fatty acids oxidized differed between groups. The relative contribution of systemic plasma fatty acids to total fat oxidation decreased, and the relative contribution of nonplasma fatty acids to total fat oxidation increased with increasing adiposity. Presumably, systemic plasma fatty acids were derived primarily from lipolysis of adipose tissue TG, and nonplasma fatty acids were derived primarily from lipolysis of intramuscular, and possibly plasma and intra-abdominal, TG. It is likely that the increased use of nonplasma fatty acids in our obese men was caused by the lesser availability and uptake of plasma fatty acids. Studies conducted in isolated skeletal muscle demonstrate a reciprocal relationship between the oxidation of plasma fatty acids and fatty acids derived from intramuscular TG during exercise (8, 9). The greater reliance in our obese than in our lean subjects on nonplasma fatty acids as a fuel during exercise is consistent with previous reports that evaluated substrate oxidation during exercise (16, 23, 27).

In summary, the results of the present study demonstrate an inverse relationship between relative body fat mass and the exercise-induced increase in fatty acid $R_a$. It is likely that the attenuated lipolytic response to exercise in overweight and obese men was caused by a blunted increase in epinephrine secretion and a concomitant reduction in adipose tissue lipolytic response to circulating catecholamines. The limited availability of systemic plasma fatty acids as a fuel in overweight and obese men was associated with a compensatory increase in the oxidation of nonplasmatic fatty acids, so the rate of total fat oxidation during exercise was similar among groups.

ACKNOWLEDGMENTS

We thank the nursing staff of the General Clinical Research Center for their help in performing the studies, Freida Custudio and Junyoung Kwon for their technical assistance, and the study subjects for their participation.

GRANTS

This study was supported by National Institutes of Health Grants HD-01459, DK-37948, RR-00954 (Biomedical Mass Spectrometry Resource), RR-00036 (General Clinical Research Center), and DK-56341 (Clinical Nutrition Research Unit).

REFERENCES

LIPID KINETICS DURING EXERCISE

E361


