Correlations Between Fatty Acid and Glucose Metabolism

Potential Explanation of Insulin Resistance of Puberty

Silva A. Arslanian and Satish C. Kalhan

In vivo resistance to the action of insulin on glucose uptake has been documented during puberty. To test the hypothesis that the glucose-fatty acid cycle, as proposed by Randle et al. (Randle PJ, Garland PB, Hales CN, Newsholme EA: The glucose-fatty acid cycle: its role in insulin sensitivity and the metabolic disturbances of diabetes mellitus. Lancet 1:785–789, 1963), may be responsible for this phenomenon, we studied nine prepubertal (Tanner I), nine pubertal (Tanner II-IV), and five young adult healthy subjects. The rate of lipolysis was measured with [d-5]glycerol tracer during basal state and during a stepwise hyperinsulinemic (10 and 40 mU • m⁻² • min⁻¹) euglycemic clamp. The rates of insulin-stimulated glucose disposal (Rd) were measured during the clamp, whereas glucose and fat oxidation were measured by indirect respiratory calorimetry. Basal glycerol rate of appearance (Ra; lipolysis) and fat oxidation were similar between prepubertal and pubertal subjects but higher than adults when the data were expressed per kilogram body weight or per kilogram fat-free mass (FFM; glycerol Rd: 2.5 ± 0.2, 2.6 ± 0.2 vs. 1.6 ± 0.2 μmol • min⁻¹ • kg FFM⁻¹, P < 0.05; fat oxidation: 4.4 ± 0.6, 4.8 ± 0.3 vs. 3.2 ± 0.6 μmol • min⁻¹ • kg FFM⁻¹, P < 0.05). However, when expressed for total body, glycerol Ra and fat oxidation were higher in pubertal versus prepubertal and adult subjects. Insulin-like growth factor I (IGF-I) levels correlated with total body lipolysis (r = 0.52, P = 0.006) and with total lipid oxidation (r = 0.44, P = 0.016) at baseline. During the low-rate insulin clamp, glycerol Ra and fat oxidation were higher in pubertal versus prepubertal adult subjects (1.5 ± 0.2 vs. 0.9 ± 0.1 μmol • min⁻¹ • kg FFM⁻¹, P = 0.04, and 3.7 ± 0.4 vs. 2.2 ± 0.4 μmol • min⁻¹ • kg FFM⁻¹, P = 0.03, respectively). During the high-rate insulin clamp, fat oxidation was significantly higher in pubertal (1.7 ± 0.3) versus prepubertal (0.7 ± 0.2) versus adult subjects (0.4 ± 0.2 μmol • min⁻¹ • kg FFM⁻¹), and IGF-I levels correlated positively with total body lipid oxidation (r = 0.72, P < 0.001). Insulin-stimulated total and nonoxidative Rd were significantly lower in pubertal subjects compared with prepubertal and adult subjects (total Rd:

57.7 ± 3.6 vs. 75.8 ± 2.8 vs. 70.0 ± 4.6 μmol • min⁻¹ • kg FFM⁻¹; nonoxidative Rd, 34.9 ± 3.9 vs. 44.6 ± 1.4 vs. 48.9 ± 3.9 μmol • min⁻¹ • kg FFM⁻¹). Fat oxidation correlated inversely with glucose oxidation, with nonoxidative Rd, and with total Rd. Moreover, the percentage of decrease in lipid oxidation during the clamp correlated with the percentage of increase in carbohydrate oxidation (r = 0.55, P = 0.004). In summary, insulin action in suppressing lipid oxidation and stimulating glucose Rd is decreased during puberty. These data suggest that increased lipid oxidation during puberty may contribute to pubertal insulin resistance at high physiological levels of insulinaemia. The mechanisms for these findings remain to be determined but could be influenced by elevated growth hormone and IGF-I levels in puberty. Diabetes 43:908–914, 1994.

Indirect evidence indicates that, during puberty, there is resistance to the action of insulin on glucose disposal (Rd) that manifests itself with hyperinsulinaemia in nondiabetic subjects (1) and with increasing insulin requirements and deteriorating glycemic control in diabetic adolescents (2). Direct evidence for pubertal insulin resistance, however, became apparent when Amiel et al. (3) demonstrated that insulin-stimulated glucose Rd was significantly lower in children at Tanner stages II-IV compared with those at Tanner stage I and compared with adults (3), and similar findings were reported by others (4). The causes and mechanisms of insulin resistance during puberty are not clear. It is not known which of the metabolic pathways of glucose disposal is affected. Whether the decrease in insulin-stimulated Rd during puberty results from a reduction in oxidative or nonoxidative Rd or both remains to be answered. We hypothesized that growth hormone (GH) secretion, which is known to increase during puberty (5–6), leads to increased lipolysis and increased free fatty acid (FFA) oxidation. This, according to Randle et al. (7), would compete with glucose oxidation, resulting in decreased glucose uptake and insulin resistance.

To test this hypothesis, we compared rates of lipolysis, insulin-mediated glucose Rd, glucose oxidation, and nonoxidative Rd in prepubertal, pubertal, and normal adult healthy volunteers.

RESEARCH DESIGN AND METHODS

Nine prepubertal children, nine pubertal adolescents, and five young adults were studied in the General Clinical Research Center (GCRC) at Children's Hospital of Pittsburgh. All studies were approved by the Children's Hospital of Pittsburgh. All studies were approved by the...
Clinical and biochemical characteristics of study subjects

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<th>Tanner II-IV</th>
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<td>57.6 ± 6.2</td>
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<td>IGFI (ng/ml)</td>
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<td>566 ± 46*</td>
<td>345 ± 35</td>
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<tr>
<td>Testosterone (nM)</td>
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<td>15.6 ± 1.8*</td>
<td>28.8 ± 3.5</td>
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<tr>
<td>Estradiol (pM)</td>
<td>&lt;18</td>
<td>62 ± 15</td>
<td>250 ± 143</td>
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*P < 0.05 pubertal vs. prepubertal vs. adult subjects.

**TABLE 1**

Biochemical measurements. Plasma glucose was measured by the glucose oxidase method with the use of a YSI glucose analyzer (Yellow Springs Instruments, Yellow Springs, OH). Plasma insulin was analyzed by radioimmunoassay (RIA; 12). FFAs were quantitated by an enzymatic colorimetric method with the use of the Wako NEFA C test kit (Wako, Richmond, VA). Glycerol was measured by microfluorometric assay (13). Glycohemoglobin was measured by high-performance liquid chromatography (Diamat, Bio-Rad, Hercules, CA), normal range being 4.9–7.3%. IGFI was measured by RIA after acid-ethanol extraction (Nichols, San Juan Capistrano, CA). Urinary nitrogen was measured by the Kjeldahl method (14).

Deuterium enrichment of glycerol in the plasma was determined according to previously described methods (15). Plasma samples were deproteinized with barium hydroxide and zinc sulfate. The supernatant was purified through a column of mixed-bed anion and cation exchange chromatography. Acetate derivatives of glycerol were prepared by adding pyridine and acetic anhydride to the dried eluted samples. Derivatized samples were analyzed for 2H enrichment on the Hewlett Packard GC-MS system (5985A; Hewlett Packard, Palo Alto, CA), as reported previously (15). Selected ion monitoring software was used to monitor charge-to-mass ratio (m/z) (145) and m/z (146 + 3), representing unlabeled and 2H-labeled glycerol, respectively. Standard curves of known enrichments of glycerol were performed with each assay.

The 3O enrichment of expired CO2 distillate from breath was measured with the use of an isotope ratio mass spectrometer (11).

**Calculations.** Total-body water was calculated from 2H enrichment of expired CO2 (11,16). Fat-free mass (FFM) and fat mass were calculated from total-body water measurements on the basis of the concept that body water is estimated relative to a fixed fraction (73.2%) of the FFM (17). Body composition. Body composition was determined by the hyperinsulinemic-euglycemic clamp (10) in conjunction with indirect calorimetry. Intravenous crystalline insulin (Humulin, Lilly, Indianapolis, IN) was infused at a constant rate of 10 mU and 40 mU·m⁻²·min⁻¹, each for 2 h. The lower insulin infusion rate was chosen to study suppression of lipolysis. The higher insulin infusion rate was used to assess total-body lipolysis and was normalized per kilogram FFM to the rate of appearance (Ra) of endogenous glycerol in plasma was calculated during the last 30 min of the clamp experiment after completion of the basal studies. Steady-state plateau of FFM was determined by multiplying glycerol R of three, because when a triglyceride molecule is hydrolyzed, one glycerol molecule and three fatty-acid molecules are produced.

**Insulin-stimulated glucose Ra.** This was calculated during the last 30 min of the steady-state condition. Under steady-state conditions of euglycemia, the rate of exogenous glucose infusion was assumed to be equal to the rate of insulin-stimulated glucose disposal. Insulin at this dose level inhibits hepatic glucose production in prepubertal and pubertal subjects (20). Because the skeletal muscle or the FFM is the predominant site for insulin-mediated glucose uptake, and the FFM is the predominant site for insulin-mediated glucose disposal, the oxygen consumption was calculated as micromoles per minute per kilogram FFM and as micromoles per minute per kilogram body weight.

Basal and insulin-stimulated carbohydrate oxidation rates and lipid oxidation were calculated from the indirect calorimetric data by averaging the data for 30 min before the beginning of the insulin infusion and for the last 30 min during insulin infusion. The protein oxidation rate was measured from urinary nitrogen excretion, and the carbohydrate and lipid oxidation rates were determined according to the formulas of Prayn (22). Nonoxidative R was estimated by subtracting the rate of glucose oxidation from the total-body insulin-stimulated Ra during the last 30 min of the clamp.

**Statistical analysis.** Statistical analyses were performed using analysis of variance for three group comparisons, and unpaired two-tailed Student's t test with correction for multiple comparisons. To evaluate univariate relationships, least-squares regression analysis was applied. Data are presented as means ± SE. P < 0.05 was considered statistically significant.

**RESULTS**

Body composition. FFM showed a significant increase from prepuberty to adulthood (Table 1). No differences were noted among the groups in percentage of body fat and total body fat.
Basal metabolic data. No differences were noted between pubertal vs. adult subjects; tP = 0.02 pubertal vs. adult subjects; tP < 0.05 pubertal vs. prepubertal vs. adult subjects.

Comparison between pubertal and prepubertal children

**Basal metabolic data.** No differences were noted between the two groups in fasting plasma glucose, insulin, glycerol, and FFA levels. Basal glycerol \( R_a \) (lipolysis), FFA \( R_a \), and lipid oxidation were similar between the two groups when the data were expressed per kilogram FFM (Table 2) or per kilogram body weight. However, when data were expressed for total body, glycerol \( R_a \), FFA \( R_a \), and lipid oxidation were higher in pubertal compared with prepubertal subjects (glycerol \( R_a \) 115 ± 10 vs. 51 ± 11 \( \mu \)mol/min, \( P = 0.03 \); lipid oxidation: 206 ± 22 vs. 136 ± 17 \( \mu \)mol/min, \( P = 0.02 \)). Basal glucose oxidation was higher in prepubertal children compared with pubertal adolescents per kilogram FFM (Table 2) and per kilogram body weight (16.2 ± 1.6 vs. 12.4 ± 0.7 \( \mu \)mol • min • kg • FFM, \( P = 0.05 \)), but similar when data were expressed for total body (592 ± 51 and 635 ± 47 \( \mu \)mol/min).

**Insulin suppression of lipolysis and lipid oxidation.**

During the low-rate insulin infusion (10 mU • m • min), no differences were noted between pubertal and prepubertal subjects in regard to steady-state glucose (5.4 ± 1 vs. 5.5 ± 0.2 mM), insulin (227 ± 20 vs. 240 ± 18 \( \mu \)U/mL), glyceral (15 ± 1 vs. 14 ± 2 \( \mu \)M), and FFA (203 ± 23 vs. 256 ± 38 \( \mu \)M) levels. Glycerol \( R_a \), FFA \( R_a \), and lipid oxidation were similar between the two groups (Fig. 1), whether data were expressed per kilogram body weight or per kilogram FFM. However, when expressed for total body, lipid oxidation was higher in pubertal compared with prepubertal subjects (166 ± 22 vs. 104 ± 17 \( \mu \)mol/min, \( P = 0.05 \)).

During the high-rate insulin infusion (40 mU • m • min), no differences were noted between pubertal and prepubertal groups in steady-state plasma glucose (5.6 ± 0.1 vs. 5.6 ± 0.1 mM), insulin (705 ± 57 vs. 700 ± 42 \( \mu \)U/mL), glyceral (11 ± 1 vs. 10 ± 2 \( \mu \)M), and FFA (100 ± 11 vs. 139 ± 24 \( \mu \)M) concentrations. Glycerol \( R_a \) was similar between the two groups (Fig. 1) regardless of the method of expression. The percentage of suppression in lipolysis from baseline was similar between the groups. Lipid oxidation, however, was higher in pubertal compared with prepubertal children per kilogram FFM (1.7 ± 0.3 vs. 0.7 ± 0.2 \( \mu \)mol • min • kg • FFM, \( P = 0.01 \)), per kilogram body weight (1.4 ± 0.2 vs. 0.6 ± 0.2 \( \mu \)mol • min • kg • FFM, \( P = 0.009 \)), and per total body (77 ± 12 vs. 21 ± 8 \( \mu \)mol/min, \( P = 0.002 \)). The percentage of suppression from baseline in lipid oxidation was lower in pubertal versus prepubertal subjects (63 ± 5 vs. 83 ± 7%, \( P = 0.03 \)). These findings are indicative of decreased insulin action in suppressing lipid oxidation during puberty.

**Insulin stimulation of glucose \( R_g \).** During the high-rate insulin infusion clamp, insulin-stimulated \( R_g \) was lower in pubertal versus prepubertal subjects per kilogram FFM (57.7 ± 3.6 vs. 75.8 ± 2.8 \( \mu \)mol • min • kg • FFM, \( P = 0.002 \); Fig. 2) and per kilogram body weight (48.7 ± 3.8 vs. 63.7 ± 4.1 \( \mu \)mol • kg • FFM, \( P = 0.02 \)). Similarly, oxidative and non-oxidative \( R_g \)s were lower in pubertal versus prepubertal subjects (oxidative: 22.2 ± 1.6 vs. 31.2 ± 2.2 \( \mu \)mol • min • kg • FFM, \( P = 0.007 \); nonoxidative: 34.9 ± 3.9 vs. 44.6 ± 1.4 \( \mu \)mol • min • kg • FFM, \( P = 0.04 \)). Similar results were obtained when the data were expressed per kilogram body weight.

**Comparison between pubertal and adult subjects.**

**Basal metabolic data.** No differences were noted between pubertal and adult subjects in fasting glucose, insulin, FFA, and glyceral levels. Basal glycerol \( R_a \) was higher in pubertal versus adult subjects per kilogram FFM (2.6 ± 0.2 vs. 1.6 ± 0.2 \( \mu \)mol • min • kg • FFM, \( P = 0.008 \); Table 2), per kilogram body weight (2.2 ± 0.2 vs. 1.4 ± 0.2 \( \mu \)mol • kg • FFM • min • kg • FFM, \( P = 0.002 \)), and per total body (2.2 ± 0.2 vs. 1.4 ± 0.2 \( \mu \)mol • kg • FFM • min • kg • FFM, \( P = 0.002 \)).
Insulin suppression of lipolysis and lipid oxidation.

During the low-rate insulin infusion, no differences were noted between the steady-state glucose, insulin, and glycerol of the pubertal and adult subjects. However, FFA tended to be higher in pubertal subjects (256 ± 38 vs. 162 ± 35 μM, P = 0.06). During this step, glycerol R_{al} FFA R_{al}, and lipid oxidation were higher in pubertal versus adult subjects (glycerol R_{al} 1.5 ± 0.2 vs. 0.9 ± 0.1 μmol·min^{-1}·kg FFMM^{-1}, P = 0.04; lipid oxidation 3.7 ± 0.4 vs. 2.2 ± 0.4 μmol·min^{-1}·kg FFMM^{-1}, P = 0.03; Fig. 1). Results were similar when data were expressed per kilogram body weight (data not shown). When the data were expressed per total body, the results showed a similar tendency but did not reach a statistical significance.

During the 40 mU·m^{-2}·min^{-1} insulin clamp, pubertal and adult subjects had similar steady-state plasma glucose (5.6 ± 0.1 vs. 5.6 ± 0.1 mM), insulin (700 ± 42 vs. 673 ± 37 pm), and glycerol levels (10 ± 2 vs. 8 ± 1 μM) but higher FFA levels (139 ± 24 vs. 74 ± 9 μM, P = 0.01). Glycerol R_{al} during this step was higher in pubertal versus adult subjects when expressed per kilogram FFMM (1.2 ± 0.2 vs. 0.7 ± 0.1 μmol·min^{-1}·kg FFMM^{-1}, P = 0.06; Fig. 1) and per kilogram body weight but not per total body. However, the percentage of suppression of glycerol R_{al} with insulin was similar between pubertal (54%) and adult subjects (56%). Lipid oxidation was higher in pubertal versus adult subjects per kilogram FFMM (Fig. 1), per kilogram body weight (1.4 ± 0.2 vs. 0.5 ± 0.1 μmol·kg^{-1}·min^{-1}, P = 0.009), and per total body (77 ± 12 vs. 30 ± 11 μmol/min, P = 0.02). Percentage of suppression of baseline lipid oxidation with insulin was lower in pubertal versus adult subjects (63 ± 5 vs. 86 ± 5%, P = 0.01), indicating impaired insulin action in suppressing lipid oxidation in pubertal subjects compared with adults.

Insulin stimulation of glucose R_{al}.

During the high-rate insulin infusion clamp, insulin-stimulated R_{al} was lower in pubertal versus adult subjects per kilogram FFMM (Fig. 2) and per kilogram body weight (48.7 ± 3.8 vs. 60.5 ± 5.7 μmol·kg^{-1}·min^{-1}, P = 0.05). Oxidative R_{al} was similar between the two groups (Fig. 2) regardless of how the data were expressed. However, the percentage increase from baseline in glucose oxidation with insulin was significantly lower in pubertal versus adult subjects (56 ± 10 vs. 130 ± 18%, P = 0.001), indicating decreased insulin action in stimulating glucose oxidation during puberty. Nonoxidative R_{al} was lower in pubertal versus adult subjects per kilogram FFMM (Fig. 2) and per kilogram body weight (29.4 ± 3.8 vs. 42.4 ± 4.5 μmol·kg^{-1}·min^{-1}, P = 0.05).

Correlations.

At baseline, IGF-I levels correlated with total-body lipolysis (r = 0.52, P = 0.006) and with total lipid oxidation (r = 0.44, P = 0.016; Fig. 3), suggesting a role for GH in baseline lipolysis and lipid oxidation.

During the high-rate hyperinsulinemic-euglycemic clamp, insulin-like growth factor-I (IGF-I) levels correlated positively with total-body lipid oxidation (r = 0.72, P < 0.001; Fig. 3). When data were expressed per kilogram FFMM, IGF-I correlated positively with fat oxidation (r = 0.56, P = 0.003) and negatively with glucose oxidation (r = -0.52, P = 0.007) and R_{al} (r = -0.59, P = 0.004). Lipid oxidation correlated inversely with glucose oxidation (r = -0.35, P = 0.046), with nonoxidative R_{al} (r = -0.47, P = 0.014), and with total R_{al} (r = -0.59, P = 0.002; Fig. 4). However, the correlation of fat versus glucose oxidation was improved when the data were expressed as the percentage of decrement in fat oxidation versus the percentage of increment in glucose oxidation (r = -0.55, P = 0.004) to correct for individual differences of baseline values. The percentage of decrease in fat oxidation during hyperinsulinemia correlated with R_{al} (r = 0.56, P = 0.003) and nonoxidative R_{al} (r = 0.40, P = 0.03).

DISCUSSION

This study tested the hypothesis that increased lipolysis and increased lipid oxidation during puberty is responsible for the decreased insulin-stimulated glucose R_{al} and/or insulin resistance or observed during puberty. A cross-sectional evaluation of three groups of healthy subjects (prepubertal children, pubertal adolescents, and young adults) revealed the following: 1) total-body lipolysis and lipid oxidation are higher in pubertal versus prepubertal and adult subjects and correlate positively with IGF-I levels; 2) insulin action in suppressing fat oxidation is decreased during puberty, but insulin action in suppressing lipolysis is similar to prepubertal and adult subjects; and 3) insulin action in stimulating oxidative and nonoxidative glucose R_{al} is decreased during puberty compared with prepuberty and young adulthood.

We hypothesized that lipolysis would be increased in pubertal subjects because of the known increase in GH secretion during puberty (5–6). When lipolysis is expressed for the whole body in micromoles per minute, pubertal subjects have the highest rate of lipolysis. Although we did not measure serial GH levels, IGF-I levels showed a positive correlation with rates of lipolysis, suggesting a role for GH in baseline lipolysis. When lipolysis is expressed per FFM or per kilogram body weight, basal glycerol R_{al} becomes com-
Fig. 3. Relationship of IGF-I to whole-body lipolysis and lipid oxidation at baseline and during the 40 mU·m⁻²·min⁻¹ insulin-clamp in prepubertal, pubertal, and adult subjects. One prepubertal subject declined the clamp after completion of baseline evaluation. O, Tanner I; C, Tanner II-IV; A, adult. For A, r = 0.72, P < 0.001; for B, r = 0.44, P = 0.016; for C, r = 0.52, P = 0.006.

Fig. 4. Relationship of fat oxidation to insulin-stimulated total, oxidative, and nonoxidative glucose disposal during the 40 mU·m⁻²·min⁻¹ insulin-clamp in prepubertal, pubertal, and adult subjects. O, Tanner I; C, Tanner II-IV; A, adult. For A, r = -0.59, P = 0.002; for B, r = -0.47, P = 0.014; for C, r = -0.35, P = 0.046.

Physiological requirements for energy substrates necessary for growth. Regarding peripheral insulin resistance, our data agree with previous reports demonstrating lower insulin-stimulated glucose Rₜ during puberty in the hyperinsulinemic-euglycemic clamp (3-4). However, this study extends the past observations further to describe the pathways and the possible mechanisms responsible for peripheral insulin resistance during puberty. The new finding is that, during hyperinsulinemia, rates of lipid oxidation are higher during puberty compared with prepuberty and young adulthood. Furthermore, IGF-I levels show a strong relationship with...
lipid oxidation rates (Fig. 3), pointing to a possible role for GH in the elevated lipid oxidation rates of puberty.

When pubertal subjects are compared with prepubertal ones, insulin-stimulated $R_o$ is 25% lower. This decrement is equally distributed between the oxidative (46%) and nonoxidative pathways (54%). When pubertal subjects are compared with adults, however, the 18%-lower insulin-stimulated $R_o$ appears to be due to lower rates of nonoxidative $R_n$ with no difference in glucose oxidation (Fig. 2). However, pubertal subjects had 1.6-fold higher basal levels of glucose oxidation than did adults. During hyperinsulinemia, the increase in glucose oxidation from baseline was significantly lower in pubertal (56%) versus adult (131%) subjects, which indicates decreased insulin action in stimulating glucose oxidation during puberty.

The positive correlation between an insulin-stimulated percentage of decrement in fat oxidation and percentage of increment in glucose oxidation may suggest a role for the Randle cycle in pubertal insulin resistance (7). Several groups have demonstrated that elevation of plasma FFA induced by infusion of triglycerides or heparin or both inhibited carbohydrate oxidation and $R_o$ in normal healthy subjects (26-28). Furthermore, it was found that the lipid-heparin infusion not only promptly replaced carbohydrates as fuel for oxidation but also inhibited glucose uptake by interfering with muscle glycogen formation with a concomitant decrease in muscle glycogen synthase activity (29). Although correlations do not imply causation, the higher lipid oxidation in pubertal subjects during hyperinsulinemia possibly is responsible for the decreased glucose oxidation and disposal and hence insulin resistance of puberty.

We found no differences among the groups with regard to insulin action in suppressing lipolysis. In all three groups, equivalent suppression was observed in glycerol $R_o$ around 43% with the low-dose insulin and around 58% with the high-dose insulin. Even though suppression of lipolysis was similar among the groups, suppression of lipid oxidation was lowest in pubertal subjects, with significant correlation with IGFI levels (Fig. 3). Studies in dogs and in humans have shown that GH increases lipid oxidation and decreases glucose $R_o$ (30-31). Moreover, GH has been shown to inhibit FFA re-esterification, thus increasing FFA availability for oxidation (32). In pubertal subjects, FFA re-esterification during hyperinsulinemia possibly was 45% lower than prepubertal values. Thus, increased GH secretion during puberty remains a likely candidate for the elevated rates of lipid oxidation.

Our observation of a gradation in basal glucose oxidation rates from highest values in prepubertal children to lowest values in adults agrees with the increased glucose turnover rates reported in childhood (33). A similar trend was observed in a study comparing obese and lean children, where the authors found a negative correlation between basal glucose oxidation and pubertal stages in lean but not in obese children (34).

In summary, we have demonstrated that lipid oxidation is increased during puberty and insulin-stimulated glucose $R_o$ is decreased. Because of the nature of our study, causal conclusions cannot be drawn from the demonstrated correlations. However, on the basis of the above-discussed literature, our data are consistent with the following hypothesis: Increased lipid oxidation during puberty, regulated possibly by increased GH secretion, may be responsible for decreased glucose disposal and thus insulin resistance, as suggested by Randle et al. (7). Investigations on potential sites and mechanisms of pubertal insulin resistance may provide important insights into the metabolic impact of insulin resistance on growth.

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REFERENCES

1. Smith CP, Williams AJK, Thomas JM, Archibald HR, Algar VB, Bottazzo GF, Gale EAM, Savage MO: The pattern of basal and stimulated insulin responses to intravenous glucose in first degree relatives of type 1 (insulin-dependent) diabetic children and unrelated adults aged 5 to 60 years. Diabetologia 31:430-434, 1988
23. Nuijten N, Campbell P, Kennedy FP, Miles M, Gerich JE: Insulin dose-response characteristics for suppression of glycerol release and
conversion to glucose in humans. Diabetes 35:1326–1331, 1986