Synergistic Effects of Testosterone and Growth Hormone on Protein Metabolism and Body Composition in Prepubertal Boys

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During human puberty there is a substantial increase in growth hormone (GH) and sex steroidal hormone concentrations, as well as in GH production rates and insulin-like growth factor-I (IGF-I) These studies were designed to investigate some of the interactions of testosterone (T) and GH in the metabolic changes of puberty. Ten boys with severe GH deficiency (GHD) were studied (mean age, 12.5 ± 0.5 years) using stable isotope infusions, indirect calorimetry, and body composition analysis. After the baseline study, they received 2 doses of T enanthate (50 to 75 mg, intramuscular [IM]), and they were studied again 4 weeks later. The boys were then begun on daily subcutaneous (SC) GH (0.3 mg/kg/wk), while T therapy was continued for another 4 weeks and the studies repeated a third time. The treatment order was randomized. Protein oxidation rates decreased after T alone (−28%, *P* < .01), decreasing further after combined T/GH treatment (−36% *v* baseline, *P* < .01). The nonoxidative leucine disposal (NOLD), a measure of whole body protein synthesis, increased significantly after combined T/GH regardless of treatment order. The combination of T/GH also resulted in greater changes in body composition than T alone, with comparable decreases in %FM and corresponding increases in fat free mass (FFM). Measures of carbohydrate (CHO) metabolism, including glucose production and oxidation rates, were unaffected by either T or T/GH combination. Plasma IGF-I concentrations increased after T treatment and even more after T/GH combination, regardless of the treatment order. In conclusion GH and T are synergistic on whole body protein anabolism and body composition in males, even at a young age, but the positive effects of T on protein anabolism and body composition appear to need a basal amount of GH for those effects to be observed. GH and T both potentiate the development of the full body composition and metabolic changes of puberty.

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A WEALTH OF available data provide ample evidence of the potent anabolic effects of growth hormone (GH) in GH-deficient individuals. Whole body protein synthesis rates increase after GH treatment of healthy volunteers and hypopituitary adults[1,2] and muscle protein synthesis increases after even 4 hours of administration to healthy controls.[3,4] Improved body composition patterns, ie, increased FFM and decreased adiposity, are typically observed after GH therapy both in adults and children even after 10 weeks of treatment and are some of the key reasons for the replacement of the GH-deficient adult with GH.[5-7] Androgenic steroids are also potent anabolic hormones. There are increased whole body and muscle protein synthesis rates, as well as increased muscle strength observed after short-term treatment with testosterone (T) in boys and adult males,[8-10] and reciprocal decreases in these measures with T suppression using a gonadotropin-releasing hormone (GnRH) analogue.[11] T suppression is associated with increased adiposity and decreased lipid oxidation rates, despite normal GH and insulin-like growth factor-I (IGF-I) production,[11,12] and T appears to be needed for the full lipolytic effect of GH in the male rat.[13]

During male puberty there is a marked increase of both of these potent anabolic hormones, and although GH is critically important for linear growth and T for virilization, both are needed to achieve the body composition changes that transform the adolescent to the fully mature adult.[14] However, the specific interactions of gonadal hormones and GH on the intermediate metabolism of substrates are not fully characterized. To explore further the metabolic effects of these 2 hormones, we studied prepubertal boys with GH deficiency (GHD) treated with T alone and T and GH in combination using stable isotope techniques, indirect calorimetry, and body composition tools. Marked synergy of effects on protein metabolism and body composition changes was observed.

MATERIALS AND METHODS

Study Subjects

These studies were approved by the Nemours Children's Clinic Clinical Research and Review Committee and the Baptist Medical Center Institutional Review Committee. Ten prepubertal boys Tanner stage I (genital and pubic hair) with clinical and biochemical evidence of GHD were recruited after informed written consent and child's assent. GHD was defined as growth retardation, poor growth velocity, and peak GH responses to secretagogues <10 ng/mL. Their clinical characteristics are summarized in Table 1. Most subjects had isolated GHD, 3 had organic GHD on stable thyroid replacements.

Experimental Design

Subjects consumed a weight maintenance diet of ~ 55Kcal/kg for 3 days before admission to the Wolfson Children's Hospital clinical research center (CRC). The afternoon preceding the baseline study (D1), body composition assessment was obtained via bioelectrical impedance analysis (BIA) using a Quantum BIA 101Q system (RLJ Systems, Clinton Township, MI). Electrodes were placed as per manufacturer's specifications on the dorsum of the right hand and foot. Subjects were fed dinner at 6 PM, fed a snack at 9 PM, and then remained fasting until the following day at 12 PM. After application of a local

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Table 1. Clinical Characteristics of Study Subjects

Age (yr)	12.5 ± 0.5
HT SDS	−2.7 ± 0.3
BMI (kg/m^2)	18.1 ± 0.8
Peak GH to stimuli (μg/L)	4.2 ± 0.7
Bone age (yr)	10.9 ± 0.4

anesthetic cream (EMLA), (Astra Zeneca, Wilmington, DE) 2 intravenous (IV) needles were placed, one in a dorsal hand vein for frequent blood sampling and the other in a contralateral antecubital vein for isotopic tracer infusions. The hand was kept warm for arterialized sampling.[15] At 8 AM (time 0), primed dose constant infusions of L-[1-^{13}C]-leucine (4.5 μmol/kg; 0.07 μmol/kg · min) and [6,6^{2}H$_2$] glucose (33 μmol/kg; 0.33 μmol/kg · min) were begun and continued for the next 240 minutes. Frequent blood sampling was obtained for isotopic tracer enrichments in plasma, as well as hormones, growth factors, and glucose concentrations. Urine nitrogen determinations were also obtained as described below. Serial breath collections were obtained for $^{13}CO_2$ enrichment in expired air. Indirect calorimetry was performed 3 times during the isotopic infusions using a CPX max indirect calorimeter (Medical Graphics, St. Paul, MN). After completion of isotopic tracer infusions at 12 PM, subjects were fed lunch and discharged.

After the baseline study (D1) was completed, subjects received an injection of T enanthate 50 to 75 mg intramuscularly (IM) (depending on weight) and the dose repeated 4 weeks after the first injection (group A). Subjects were then admitted for a repeat infusion study (D2), identical to that described for D1, within 3 days of the second injection. Patients were then begun on daily subcutaneous (SC) injections of recombinant human GH at a dose of 0.3 mg/kg · wk. T treatment was continued and a third injection given 5 weeks from the second. An identical infusion study was repeated for a third time (D3) within 3 days of the last T injection (5 weeks from D2). Group B had the treatment order randomized, ie, received T and GH in combination first followed by T alone.

Blood and Breath Samples

Isotopic enrichments of α-ketoisocaproic acid (^{13}-KIC) and $^2H_2$-glucose were obtained at −20, 160, 180, 200, 220, and 240 minutes. Plasma IGF-I, insulin-like growth factor-binding protein (IGFBP)-3, insulin, glucose, free T and T concentrations were measured at times 0, 120, and 240 minutes. Breath collections were obtained at −20, −15, −10, 160, 180, 200, and 220 minutes for expired labeled CO_2. A 4-hour urine collection was obtained during the tracer infusions (8 AM to 12 PM) and a small aliquot analyzed for urea nitrogen concentration. Although this was only a 4-hour, not a 24-hour urine collection, because the experimental paradigm after hormonal treatment was identical, we believe that the comparisons are valid.

Assays

The enrichments of [$^2H_2$]glucose were measured at the Nemours Biomedical Analysis Laboratory by gas chromatography/mass spectrometry (GC/MS) similar to the methodology previously described.[16-18] Plasma samples were deproteinized using acetone, the mixture was centrifuged, and the resulting supernatant evaporated to dryness under nitrogen. A pentacetate derivatization was accomplished by the addition of acetic anhydride and ethylacetate with heat. The samples were then analyzed using a HP 5890 Series II Gas Chromatograph with a J & W Scientific DB-1701 (20 m × 0.18 mm ID × 0.4 μm film thickness) column equipped with a HP 5970 Series Mass Selective Detector (EI) (Agilent Technologies, Palo Alto, CA). Single ion monitoring (SIM) was used monitoring 98 m/z for glucose and 100 m/z for [$^2H_2$]glucose peaks. The intra-assay coefficient of variation (CV) was 1.1%.

The plasma enrichment of α-ketoisocaproic acid (α-KIC) was used as an index of the intracellular enrichment of leucine (reciprocal pool model[19,20]). The enrichments of [^{13}C]KIC were measured as previously described.[21-23] In summary, plasma samples were deproteinized with hydrocholoric acid (HCl). The plasma fraction containing KIC was purified using an AG 50 strong acid cation exchange resin and the eluent alkalinized (pH 14) with sodium hydroxide (NaOH). An oxime derivative was formed by adding Hydroxylamine HCl and heat, the solution then acidified with HCl, and extracted with ethylacetate. The ethylactate was evaporated to dryness under nitrogen. Samples were then derivatized with N-methyl-n-t-butyldimethylsilyl triflouroracetamide and analyzed using the same GC/MS as above. SIM was used monitoring 316 m/z for KIC, 317 m/z for [^{13}C]KIC, and 323 m/z for [$^{13}C_7$]KIC (internal standard) peaks. The intra-assay CV was also 1.1%.

Exhaled $^{13}CO_2$ was determined using a HP 5890 Series II Gas Chromatograph with a Chrompack Hayesep Q (2.5 M × 1/8" × 2 mm) packed column equipped with a Micromass Optima Isotope Ratio Mass Spectometer (Micromass, Manchester, UK). Vacutainers containing breath samples were loaded into the Gilson Isochrom μGas Autoinjector. SIM was used monitoring 44 m/z for CO_2, and 45 m/z for $^{13}CO_2$.[24,25]. The intra-assay CV was 0.22%.

Insulin, IGF-I, IGFBP-3, T, and free T concentrations were measured at the immunochemical core laboratory at the Mayo Clinic GCRC (Rochester, MN) using commercial kits. Serum GH concentrations were measured by highly sensitive chemiluminescence assay at the University of Virginia CRC core laboratory (Charlottesville, VA). Plasma glucose levels were determined by glucose oxidase methods using a Beckman glucose analyzer (Beckman Instruments, Palo Alto, CA) at bedside. Urea nitrogen concentrations measured using a Kodak Ektakem urease method (Rochester, NY).

Calculations

The rate of appearance (Ra) of glucose, a measure of hepatic glucose output, was calculated as Ra = [(Ei-Ep)−1]F, where Ei is the isotopic enrichment of the infusate, Ep represents the enrichment of glucose in plasma and F is equal to the rate of infusion. Estimates of whole body leucine metabolism were calculated at near substrate and isotopic steady state between 180 and 240 minutes of isotopic infusions. Rates of whole body protein synthesis and degradation were determined using validated steady state equations and the reciprocal pool model.[1,19,20] The Ra of leucine (μmol/kg · min) was calculated similarly to the Ra of glucose using the mean enrichment of [^{13}C]KIC at steady state. Leucine oxidation rates were calculated using the [^{13}C]KIC enrichment and the CO_2 recovery in each individual corrected by a factor of 0.80.[9,19,20] The expired rate of $^{13}CO_2$ was determined by multiplying the $^{13}CO_2$ enrichment by the total CO_2 production. Because the subjects were fasting, we assumed that at steady state, the Ra = rate disappearance, hence the nonoxidative leucine disappearance (NOLD) = Ra − leucine oxidation rate.

Protein, glucose, and lipid substrate oxidation rates and resting energy expenditure were calculated using VO_2 and VCO_2 gas exchange rates obtained via indirect calorimetry as lipid oxidation = $1.67(VCO_2 - VO_2) + 1.92N$; glucose oxidation = $4.35VCO_2 - 3.21VO_2 - 2.87N$; protein oxidation = N × 6.25; resting energy expenditure = $[3.91(VCO_2)/10^3] + [1.1(VO_2) - 3.71N/10^3]$ where VCO_2 and VO_2 are the liters/minute of gaseous exchange obtained by calorimetry and N represents the total nitrogen excretion (g/min) measured.[26]

Body composition was measured using BIA as described by Houtkooper et al[27] for use in children using the following formula: FFM = $[0.61(\text{height})^2/R] + 0.25(\text{weight}) + 1.31$, where FFM = fat free mass and R = resistance measured by the instrumentation.

Isotopes and Drugs

L-[1-^{13}C] leucine (99% enriched, Cambridge Isotopes, Andover, MA), [6,6-$^{2}H_2$] glucose (99.7% enriched, MSD Isotopes, St Louis, MO) were determined to be sterile and pyrogen free and mixed with 0.9% nonbacteriostatic saline. T enanthate (Delatestryl, 200 mg/mL) was obtained commercially (BTG Pharmaceuticals, Iselin, NJ). GH (Nutropin, 5 mg/mL) was kindly provided by Genentech (South San Francisco, CA).

Statistical Analysis

Results are expressed as mean ± SE. Repeated measures analysis of variance (RM ANOVA) was used to determine differences between baseline studies and the 2 treatment arms of the study for all parameters tested. In this particular design, we tested for a "main effect" of treatment type (baseline, T, T/GH), a main effect of group order (A, B), and an interaction effect between the 2 main effects. If there was a significant difference in any of these 3, we further explored with post hoc comparisons. The advantage of this approach is that the problem of multiple comparisons is more efficiently handled. Significance for all statistical analyses was established at $P < .05$. Data are shown as mean ± SEM.

RESULTS

Protein Kinetics

There was a significant treatment effect on the Ra of leucine ($P = .006$), leucine oxidation ($P < .001$), and NOLD ($P < .001$). There were no significant group or group by treatment order interaction effects for any of the protein kinetics variables. Post hoc comparisons demonstrated that the Ra of leucine and NOLD were significantly different from baseline after T/GH ($P < .01$, $P < .001$), but not after T alone ($P = .754$, $P = .531$) in groups A and B, respectively. Leucine oxidation rates, however, were significantly lower from baseline after either T/GH ($P = .001$) or T alone ($P = .005$) (Fig 1).

Body Composition

Weight increased significantly after combined T/GH treatment regardless of treatment order and increased after T alone only in group B (group A baseline: 35 ± 4.5 kg; T: 35.8 ± 4.0; T/GH: 37.3 ± 4.2*; group B baseline: 33.8 ± 4.5; T/GH: 35.5 ± 5.0*; T: 34.8 ± 4.5*; [*$P < .01$ *v* baseline]). There were no changes in body mass index (BMI) except for a modest increase (4 %) after combined T and GH therapy in group B. There were differences in body composition changes after T alone depending on the treatment order, with no significant change in %FM when the T alone was administered first (not statistically significant) versus 6.4% less %FM when the T-alone arm followed the combined T/GH arm ($P = .001$ *v* baseline). After combined T/GH, there were comparable decreases in %FM regardless of treatment order (group A: ~−5.3%; group B: ~8.3% *v* baseline, $P < .01$, both) and corresponding increases in FFM (group A: ~+3.7 kg, $P < .001$; group B ~+4.3 kg%, $P < .01$) (Fig 2).

Carbohydrate Metabolism

Fasting glucose concentrations increased significantly only in group B after the administration of T/GH, however, mean serum glucose levels remained within a normal physiologic range (Table 2). Although there was a trend towards increased insulin output after combined T/GH treatment in groups A and B, it did not achieve statistical significance. No change in mean fasting insulin levels was observed after T or GH alone. Glucose production rates (glucose Ra) were measured by stable isotope methods only in group B, and they did not change after either T or combined treatment.

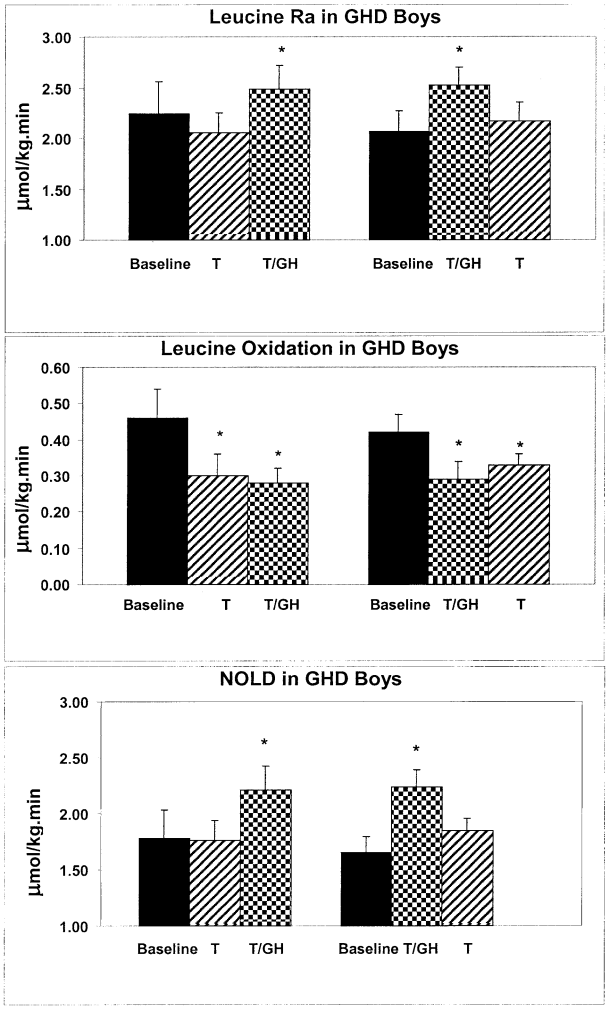

Fig 1. Rates of (A) whole body proteolysis (leucine Ra), (B) leucine oxidation, and (C) nonoxidative leucine disposal in a group of prepubertal GH-deficient boys (n = 10) at baseline, after treatment with GH for 4 weeks, followed by T and GH in combination for another 4 weeks. The left and right panels show the difference in treatment order. *$P < .05$ *v* baseline by RM ANOVA with post hoc comparisons.

Substrate Oxidation and Energy Expenditure Rates

When data for the entire cohort were averaged, the mean respiratory quotient (RQ) at baseline, after T and after T/GH was 0.8 ± 0.0. When the data of the 2 groups were combined, marked decreases in protein oxidation rates in the T and combined T/GH treatment groups were observed (baseline: 5.1 ± 0.5 kcal/kg FFM · d; T: 3.5 ± 0.4, $P = .001$; T/GH: 3.0 ± .03, $P = .0002$ *v* baseline, post hoc comparisons), irrespective of the treatment order. Carbohydrate (CHO) oxidation rates did not change significantly after T or combined T/GH (baseline: 21.5 ± 2.8 kcal/kg FFM · d; T: 19.7 ± 1.6; T/GH: 20.5 ± 2.2).

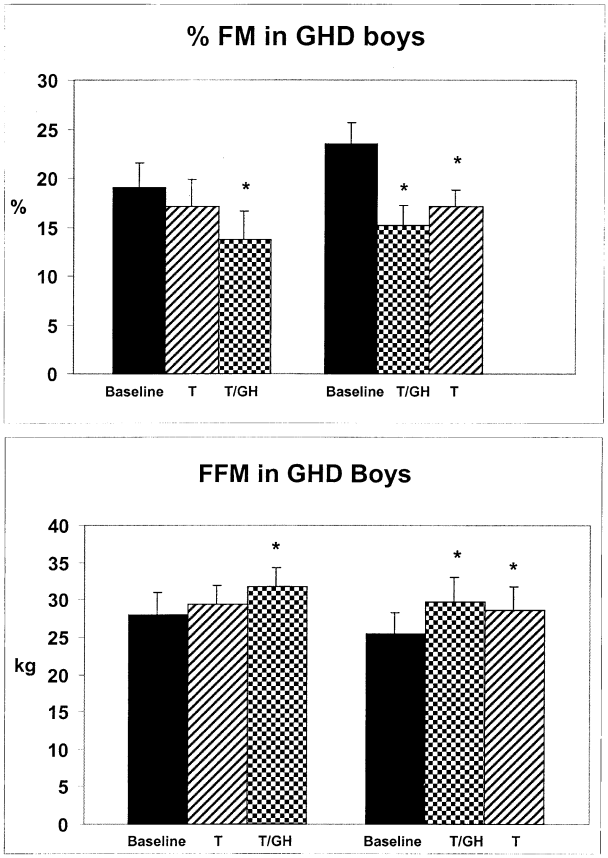

Fig 2. (A) Shows the %FM and (B) the FFM as measured by BIA in the same prepubertal boys with GHD as shown in Fig 1, at baseline, after 4 weeks of GH and 4 weeks of T/GH in random order (%FM group A baseline: 19.0% ± 2.5%; T: 17.1 ± 2.7; T/GH:13.7 ± 2.9*; group B baseline: 23.5± 2.2; T/GH: 15.2 ± 2.0*; T: 17.1 ± 1.7*; FFM group A baseline: 28.1 ± 2.9 kg; T: 29.3 ± 2.5; T/GH: 31.8 ± 2.6*; group B baseline: 25.5 ± 2.7; T/GH: 29.8 ± 3.3*; T: 28.6 ± 3.1*; *P < .01 *v* baseline).

No significant changes in lipid oxidation rates (baseline: 20.7 ± 3.2 kcal/kg FFM · d; T: 20.0 ± 1.9; GH/T 22.3 ± 3.4) or resting energy expenditure rates (baseline: 47.2 ± 3.4 kcal/kg FFM · d; T: 43.2 ± 2.2; GH/T: 45.7 ± 3.5; baseline: 45.1 ± 3.3) were observed between the treatment groups, again regardless of treatment order.

Hormones and Growth Factors

Post hoc comparisons indicated that IG-I concentrations were significantly higher than baseline after treatment with either T alone ($P = .004$) and after treatment with T/GH ($P < .001$), but with much greater concentrations after combination treatment ($P < .001$), regardless of treatment order (group A baseline: 136 ± 26 μ/L; T: 216 ± 27, $P < .04$ [1 tailed]; T/GH: 380 ± 55 μ/L, $P < .01$; group B baseline: 126 ± 29; T: 206 ± 41, $P < .01$; T/GH: 316 ± 66, $P < .01$ both *v* baseline). When analyzed in aggregate, IGFBP-3 levels did not change after T alone (baseline: 2.8 ± 0.3 mg/L; T: 2.9 ±0.3, not significant [NS]), but increased after combined T/GH treatment (3.2 ± 0.3, $P = .04$) regardless of treatment order. As expected, serum T concentrations increased significantly in both the T alone and combined T/GH treatment groups (group A baseline: 58 ± 44 ng/dL; T: 755 ± 125; T/GH: 393 ± 29, $P < .01$ both *v* baseline; group B baseline: 44 ± 9; T: 680 ± 177; T/GH 886 ± 136, $P < .02$, 0.003, respectively, both *v* baseline).

DISCUSSION

The study of prepubertal boys with GHD allows us to assess the interactions of androgens and GH in the metabolic changes of puberty. The combination of GH and T was more potent than T alone in increasing whole body protein synthesis rates regardless of the treatment order as measured by the nonoxidative leucine disposal. However, T alone did not affect protein synthesis in the hypopituitary child. This contrasts with the potent protein-anabolic effect of T monotherapy observed in prepubertal GH-sufficient boys also treated with T for 4 weeks, which we reported previously[9] and suggests that T's actions on whole body protein pools need an underlying secretory tone of normal GH production for a full effect to be observed.

We measured body composition by BIA.[27] Although this technique may overestimate the absolute lean body mass, it has been found useful in population studies in children,[28] and is tightly correlated with data generated by dual x-ray absorptiometry (DEXA).[29] In this particular paired paradigm in which the subjects are compared with each other, the ability to measure differences using the same tools, we believe, is robust.[27-30] There was a significant decrease in adiposity after treatment

Table 2. Changes in Carbohydrate Metabolism After Treatment

	Glucose (mmol/L)	Insulin (pmol/L)	Glucose Production Rates (μmol/kg · min)
Group A			
Baseline	4.84 ± 0.2	31.2 ± 6.5	NA
T	5.15 ± 0.3	32.4 ± 4.0	NA
T/GH	5.05 ± 0.2	43.8 ± 8.4	NA
Group B			
Baseline	4.85 ± 0.07	36.0 ± 9.2	17.0 ± 3.7 (3.1 ± 0.7)
T/GH	5.43 ± 0.1*	49.8 ± 12.8	15.4 ± 2.5 (2.8 ± 0.4)
T	5.30 ± 0.2	30.6 ± 6.0	16.0 ± 2.4 (2.9 ± 0.4)

NOTE. mg/kg · min in parenthesis.
Abbreviation: NA, not available.
*$P < .05$ *v* baseline.

with T in combination with GH, however, T alone reduced adiposity only when preceded by GH/T combination, but not when given by itself first. The combination treatment was more potent than T alone decreasing adiposity, regardless of treatment order. It is also possible that the decrease in adiposity, as compared with baseline, observed when the T treatment was preceded by combined T/GH treatment first (group B) is a result of the discontinuation of GH. However, the fact that T alone in group B was not able to further decrease adiposity as observed during the last visit of group A, suggest that GH is likely critical for the decrease in adiposity observed during male puberty. GH is a potent lipolytic hormone, accelerating lipolysis and increasing lipid oxidation in both experimental animals and in humans.[2,31-36] However, T also participates in lipid metabolism.[8,11,37,38] T deficiency in males, either mild as observed in elderly men, or severe, as caused by GnRH analogue treatment, results in decreased lipid oxidation, increased adiposity, and decreased lean body mass despite normal GH secretion.[8,11] In hypophysectomized rats, T treatment in conjunction with GH normalizes lipolysis rates better than GH given alone, demonstrating a synergistic effect of both hormones in fat metabolism.[13] In elderly GH-deficient patients treated with both GH and T for 6 months, the combination treatment was more potent than either compound alone in reducing subcutaneous, but not visceral fat.[39] In the present studies, on the contrary, we cannot compartmentalize the regional changes in body composition, which typically necessitate spiral abdominal computed tomography or magnetic resonance imaging. However, studies of the regional fat distribution of boys strongly suggest that GH mostly decreases abdominal adiposity in childhood,[6] and this is similar to what is observed in adult GH-deficient subjects treated with GH.[7] In recent studies in adult males treated with a GnRH analogue to induce hypogonadism, we observed that treatment with GH preserved lean body mass and %FM to pre-GnRH analogue treatment levels.[12] However, in the absence of T, GH treatment did not cause the potent lean body mass increase and decrease in adiposity typically observed in the eugonadal state.[2,12] The present data are congruent with these findings and suggest that the marked decrease in adiposity and lean body mass accrual necessary for the full body composition changes of puberty to be observed necessitates both androgens and GH.

There were no changes in measures of CHO metabolism with either treatment protocol, with no changes in glucose or insulin concentrations after either T or T/GH combination. There were no measurable changes in glucose production rates as measured by glucose Ra or in glucose oxidation rates as measured by indirect calorimetry. These results contrast with the decreased glucose oxidation observed after GH treatment of GH-deficient young adults[2,31] and mirror the findings in GnRH analogue-treated males[11] and GnRH analogue/GH-treated adults, who also did not have any changes in the oxidation of CHO.[12] Collectively, these data suggest that neither T nor GH are principal regulators of CHO utilization in vivo in this age group.

Both T alone and in combination with GH potently suppressed the oxidation of protein measured using indirect calorimetry, but more so the combination treatment, regardless of treatment order. These effects are similar to those observed when the protein oxidation was measured using the leucine tracer data. In hypogonadal men whom we treated with GH, GH also decreased the oxidation of protein, despite the absence of T.[12] In aggregate, these data support the concept that GH and T independently activate the signaling transduction signals necessary for the preservation of whole body protein, both of which result in a potent anabolic effect. Resting energy expenditure rates were much higher in these boys than those measured in young adult normal males and in GH-deficient adults,[2,12] mostly due to their high CHO oxidation rates, as compared with the adults. These rates did not change with either treatment. These data offer indirect evidence that neither GH nor T perturb the high-energy requirements of the pubertal process.

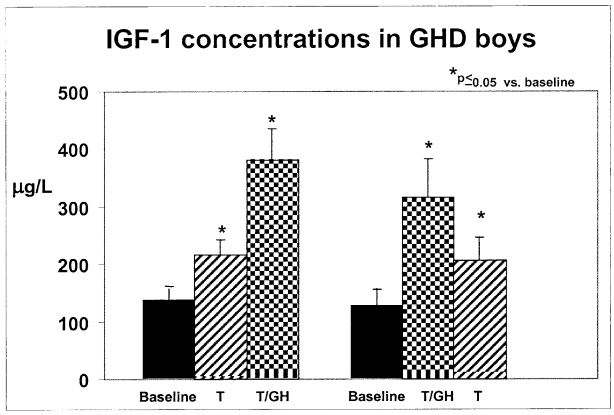

Fig 3. Plasma IGF-I concentrations at baseline, after T and T/GH in prepubertal GH-deficient boys (n = 10) with randomized treatment order.

Plasma IGF-I concentrations increased after T alone, but much more so after T and GH, again indicative of the synergy of effects of these 2 hormones. Although IGF-I increased with T alone, IGFBP-3, a major reservoir of IGF-I in plasma and an entirely GH-dependent growth factor, did not change, suggesting that, in the GH-deficient state, T's effects on the GH axis are limited.

In summary, the combination of T and GH is more potent than T alone increasing whole body protein pools, decreasing adiposity, and increasing lean body mass in prepubertal GH-deficient males, effects reflected in the changes in IGF-I and IGFBP-3 concentrations. Neither hormone affected significantly any measures of CHO metabolism, including glucose production and oxidation rates, suggesting that neither hormone is a principal regulator of CHO metabolism in this age group. In conclusion, these studies suggest that GH and T are synergistic in a variety of metabolic processes in hypopituitary males, even at a young age, including body composition and whole body protein anabolism. The marked positive effects of T on protein anabolism and body composition need a basal amount of GH for those effects to be observed. GH and T are both critically important for the development of the full set of body composition changes of puberty.

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