Interaction between Testosterone and Growth Hormone on Whole-Body Protein Anabolism Occurs in the Liver

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Context: GH and testosterone both exert protein-anabolic effects and may act synergistically. Liver and muscle are major sites of protein metabolism.

Objective: Our objective was to determine whether the site of GH and testosterone interaction on protein metabolism is primarily hepatic or extrahepatic.

Design: In this open-label randomized crossover study, the impact on whole-body protein metabolism of oral (solely hepatic testosterone exposure) and transdermal (systemic testosterone exposure) testosterone replacement in the presence or absence of GH was compared.

Patients and Intervention: Eleven hypopituitary men with GH and testosterone deficiency were randomized to 2-wk treatments with transdermal testosterone (10 mg) or oral testosterone (40 mg), with or without GH replacement (0.6 mg/d). The dose of testosterone administered orally achieves physiological portal testosterone concentrations without spillover into the systemic circulation.

Main Outcome Measures: Whole-body leucine turnover was measured, from which leucine rate of appearance (LRa), an index of protein breakdown, and leucine oxidation (Lox), a measure of irreversible protein loss, were estimated at the end of each treatment.

Results: In the absence of GH, neither transdermal nor oral testosterone affected LRa or Lox. GH therapy significantly increased LRa, an effect equally reduced by transdermal and oral testosterone administration. GH replacement alone did not significantly change Lox, whereas addition of testosterone treatment reduced Lox, with the effect not significantly different between transdermal and oral testosterone.

Conclusions: In the doses used, testosterone stimulates protein anabolism by reducing protein breakdown and oxidation only in the presence of GH. Because the net effect on protein metabolism during GH therapy is not different between systemic and solely hepatic testosterone administration, we conclude that the liver is the primary site of this hormonal interaction. **(*J Clin Endocrinol Metab* 96: 1060–1067, 2011)**

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Abbreviations: CV, Coefficient of variation; KIC, α-ketoisocaproic acid; Lox, leucine oxidation; LRa, leucine rate of appearance.

In adults with GH deficiency, muscle mass is reduced with these differences reversed upon GH replacement (1–3). Androgen deficiency also results in a reduction in muscle mass, which is normalized by testosterone replacement (4, 5). Thus, GH and testosterone both exert muscle-anabolic effect and may act synergistically. Evidence shows that both hormones are necessary to exert an optimal effect. This is exemplified in GH-deficient men in whom lean body mass remains subnormal even after adequate androgen replacement (1). Linear growth in GH-deficient children receiving GH replacement is further stimulated by androgen treatment (6), and for full androgen growth-promoting effect, GH replacement is required (7). This observation suggests that the anabolic effect of androgens may depend on the presence of GH.

In prepubertal boys with GH deficiency, testosterone and GH replacement results in a greater stimulation of whole-body protein synthesis than testosterone treatment alone (8). How GH and testosterone interact to regulate protein metabolism in adult life is poorly understood. We recently reported that in hypopituitary men, both GH and testosterone promote protein anabolism, this effect being enhanced with combined treatment (9). Thus, testosterone and GH exert independent and additive effects in regulating protein metabolism. However, the primary site of GH and testosterone interaction is unknown.

Liver and muscle are major sites where protein metabolism is regulated. We aimed to determine whether the site of GH and testosterone interaction on protein metabolism is primarily hepatic or extrahepatic. Oral delivery of testosterone exposes the liver to high portal levels of testosterone, which undergoes first-pass hepatic metabolism reducing or preventing the appearance of additional testosterone in the systemic circulation. We compared the impact on whole-body protein metabolism of testosterone administered via oral route (at doses used resulting in solely hepatic testosterone exposure) with transdermal testosterone replacement (systemic testosterone exposure) in the presence or absence of GH.

Subjects and Methods

Subjects

Eleven hypopituitary men with GH and androgen deficiency were recruited from the Endocrine Outpatient Clinic, St. Vincent's Hospital, Sydney, Australia. Study participant clinical characteristics are shown in Table 1. GH deficiency was confirmed with insulin tolerance test by peak GH response to insulin-induced hypoglycemia of less than 3 ng/ml (10). Secondary hypogonadism was confirmed by serum testosterone of less than 6 nmol/liter accompanied by low blood LH levels. The duration of hypopituitarism was at least 2 yr. Previously and throughout the study, participants received standard thyroid hormone and cortisol replacement for thyroid and adrenal deficiencies, respectively, with the doses unchanged throughout the study. Study participants discontinued testosterone replacement before commencement of the study for at least 2 wk for transdermal testosterone, 8 wk for im testosterone esters, and 6 months for depot testosterone preparations. All participants were instructed to follow their usual diet and physical activity as well as continuing their usual medications or supplements throughout the study.

The Human Research Ethics Committee of St. Vincent's Hospital approved the study. The study was conducted in accordance with the principles of the Declaration of Helsinki. All participants gave written informed consent. The study was registered with the Australian and New Zealand Clinical Trials Registry (ACTRN12605000482662).

Study design

This was an open-label, randomized two-period crossover study (Fig. 1). Participants were entered into the study after providing informed consent and sufficient washout of previous testosterone treatment. They were randomized to transdermal or oral testosterone treatments, each for 2 wk with and without GH replacement. The washout period in between the oral and transdermal testosterone treatments was 2 wk. Study participants underwent GH therapy for 3 months before commencement of

TABLE 1. Characteristics of hypogonadal GH-deficient subjects

Subject no.	Age (yr)	BMI (kg/m^2)	Diagnosis	Treatment	Hormone replacement
1	36	26.6	Craniopharyngioma	S	A, T, G
2	28	35.8	Idiopathic hypopituitarism	Nil	A, T, G, D
3	54	30.7	Pituitary macroadenoma	S	A, T, G,
4	52	29.8	Pituitary macroadenoma	S	A, T, G
5	20	28.5	Craniopharyngioma	S	G
6	26	20.6	Pituitary macroadenoma	S	A, T, G
7	73	26.2	Pituitary macroadenoma	S	A, T, G
8	73	26.3	Pituitary macroadenoma	S	A, T, G
9	49	27.8	Hypopituitarism post TBI	Nil	A, T, G, D
10	47	36.4	Hypopituitarism post cranial irradiation	Nil	A, T, G
11	53	30.0	Pituitary macroadenoma	S, X	A, T, G, D

A, Adrenal replacement; BMI, body mass index; D, deamino-8-arginine vasopressin; G, gonadal replacement; S, surgery; T, thyroid replacement; TBI, traumatic brain injury; X, radiotherapy.

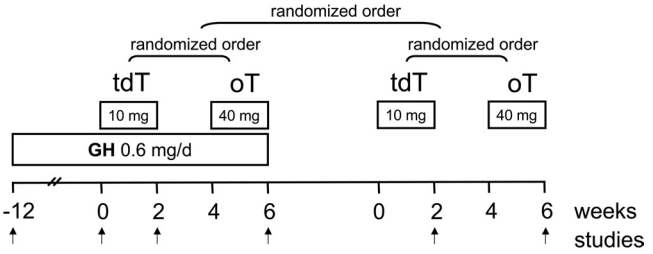

FIG. 1. Study design. Hypopituitary men were randomized to transdermal and oral testosterone treatments, each for 2 wk with or without GH replacement. The washout period in between the testosterone treatments was 2 wk. The GH sequence was randomized, and the run-in period for the GH therapy was 3 months. If the randomization sequence was such that GH therapy was started first, there was a period of GH washout for 4 wk before treatment with transdermal and oral testosterone was recommenced. The dose of transdermal testosterone was 10 mg/d, oral crystalline testosterone 40 mg/d, and GH 0.06 mg/d. The dose of oral testosterone was designed to achieve physiological portal testosterone concentrations without spillover into the systemic circulation. tdT, Transdermal testosterone; oT, oral testosterone.

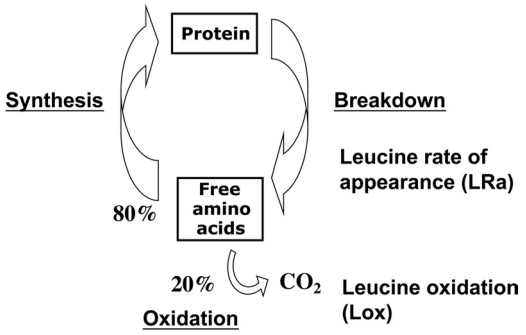

FIG. 2. Whole-body protein metabolism. In postabsorptive state, about 20% of amino acids derived from proteolysis are lost irreversibly through oxidation, and the remaining 80% are reincorporated back into protein. Leucine turnover technique is based on the principle of steady-state kinetics in which the LRa, an index of protein breakdown, equals its rate of disposal (Lox), an index of irreversible loss of protein, and leucine incorporation into protein.

testosterone treatment to achieve stable GH metabolic effects (Fig. 1). If the randomization sequence was such that GH therapy was started first, after completion of the GH phase, there was a period of GH washout for 4 wk before treatment with transdermal and oral testosterone was recommenced. The dose of transdermal testosterone (Androderm patches; Mayne Pharma Ltd., Melbourne, Australia) was 10 mg/d, and the dose of oral crystalline testosterone was 40 mg/d. Oral testosterone was prepared by Fresh Therapeutics (Sydney, Australia) as capsules filled with crystalline testosterone USP without excipients. The daily 40-mg dose of crystalline testosterone was divided into three doses taken every 8 h to achieve more constant hepatic exposure to testosterone. The dose of oral testosterone was selected based on our previous work in hypogonadal men where 40 mg/d of oral crystalline testosterone was the highest dose that did not increase systemic testosterone levels and did not reduce blood levels of SHBG, confirming the liver to be exposed to physiological testosterone levels through portal circulation without any spillover into the systemic circulation (11). GH (Humatrope; Eli Lilly Australia, West Ryde, Australia) was injected sc in a dose of 0.6 mg/d in the evening.

The effects of GH and testosterone on the following variables were studied: 1) whole-body leucine rate of appearance (LRa), an index of protein breakdown; 2) leucine oxidation (Lox), an index of oxidative loss of protein; and 3) blood levels of IGF-I, testosterone, and SHBG.

Participants were studied after an overnight fast in the early morning in the Clinical Research Facility, Garvan Institute of Medical Research. Studies were undertaken at baseline and at the end of each treatment period. At each visit, study blood samples were collected and placed on ice, and plasma was separated and stored at −80 C until analysis.

Methods

Protein turnover

Whole-body protein metabolism was measured using the leucine turnover technique. The method is based on the principle of steady-state kinetics in which the rates of appearance of substrate equals its rate of disposal. For leucine, there are two pathways of disposal: oxidation and reincorporation into protein (Fig. 2). The fractional partitioning between these two pathways of disposal is determined from the fraction of infused isotope that appears in breath. LRa and Lox were calculated as previously described (12). α-Ketoisocaproic acid (KIC) is formed when leucine undergoes transamination and is used as a surrogate marker of leucine as it more accurately reflects the intracellular environment (13).

After an overnight fast, a 0.104-mg/kg priming dose of $NaH^{13}CO_3$ was followed by a primed constant 3-h infusion of 1-[^{13}C]leucine (prime 0.5 mg/kg, infusion 0.5 mg/kg·h), as previously described (14, 15). $NaH^{13}CO_3$ and 1-[^{13}C]leucine were obtained from Cambridge Isotope Laboratories (Woburn, MA). On each visit, blood and breath samples were collected before (−10 and 0 min) and during (140, 160, and 180 min) the leucine infusion. Blood was placed on ice, and plasma was separated and stored at −80 C. KIC was extracted from plasma as described by Nissen *et al.* (16). Plasma KIC enrichment with ^{13}C was measured by gas chromatography mass spectrometry (MSD 5971A, model 5890; Hewlett-Packard Co., Palo Alto, CA). CO_2 enrichment with ^{13}C in breath samples was measured at University of Surrey (Surrey, UK) on a Delta Plus XP isotope ratio mass spectrometer fitted with a Gas Bench II inlet system (Thermo Fisher Scientific, Hemel Hempstead, UK). The coefficients of variation (CV) for LRa and Lox at the Garvan Institute are 3.5 and 6.1%, respectively.

Indirect calorimetry

For estimation of Lox, carbon dioxide production was measured by indirect calorimetry. This involved using an open-circuit ventilated-hood system (Deltatrac Metabolic Monitor; Datex Instrumentarium Corp., Helsinki, Finland), calibrated against standard gases before each study. Participants were rested on a bed for at least 30 min. A clear plastic hood was placed loosely over the subject's head for a 20-min period. Measurements were collected during two 20-min periods and averaged.

Assays

All samples for any individual were measured in the same assay run for each analyte. Serum IGF-I levels were measured by RIA after acid ethanol extraction as previously described (9, 15, 17). The CV for IGF-I were 8.3% at 14.7 nmol/liter and 7.4% at

28.6 nmol/liter. Serum testosterone, SHBG, and prostate-specific antigen (PSA) were measured by RIA using commercial assays (Immulite 2000; Siemens Medical Solution Diagnostics, Los Angeles, CA). The lowest limit of detection for testosterone assay was 0.7 nmol/liter. The interassay CV for testosterone at 3.6 and 23 nmol/liter were 9.3 and 9.0%, respectively. The CV for SHBG at 5.3 and 86.2 nmol/liter were 5.0 and 7.5%, respectively. The CV for PSA were 7.6% at 0.33 ng/ml and 5.0% at 10 ng/ml.

Statistical analysis

Treatment effects of GH, transdermal and oral testosterone alone, and combination effects were assessed using repeated-measures ANOVA followed by paired *t* tests with Bonferroni's correction. Because there were no carryover and sequence effects detected by ANOVA, data were pooled together for the treatment-effect analysis. Results are expressed as mean with SEM, and a P value < 0.05 was considered to be significant. Statistical analysis was undertaken using the statistical software package Statview version 4.5 PPC (Abacus Concepts, Inc., Berkeley, CA).

Results

The mean age of the participants was 46.4 ± 5.3 yr, and mean body mass index was 29 ± 1.3 kg/m^2. PSA levels for all participants ranged from 0.05–4.4 μg/liter. The mean weight did not significantly change throughout the study.

Testosterone

In the absence of GH treatment, transdermal testosterone administration increased the mean testosterone level significantly ($P < 0.001$; Table 2) compared with baseline, reaching the adult normal range (12–36 nmol/liter). On GH therapy, transdermal testosterone also significantly increased circulating testosterone levels ($P < 0.001$; Table 2), whereas oral testosterone did not significantly change the mean testosterone levels compared with baseline (Table 2). Blood testosterone levels on transdermal administration were significantly higher than on oral administration, regardless of GH treatment ($P < 0.01$).

IGF-I levels

In the absence of GH, neither transdermal nor oral testosterone administration significantly changed IGF-I levels (Table 2). During GH therapy, the mean IGF-I levels significantly increased compared with baseline ($P < 0.0001$). GH coadministration with transdermal and oral testosterone significantly increased circulating IGF-I compared with baseline ($P < 0.0001$; Table 2). During GH therapy, IGF-I levels significantly ($P < 0.05$) increased with oral but not transdermal testosterone treatment compared with GH therapy alone. The changes in circulating IGF-I levels between oral and transdermal testosterone during GH therapy but were not significantly different (Table 2).

SHBG levels

In the absence of GH, mean SHBG levels on transdermal testosterone administration were significantly lower compared with baseline ($P < 0.01$; Table 2), whereas on oral testosterone administration, mean SHBG levels did not significantly differ from baseline. When compared with baseline, mean SHBG levels did not significantly change during GH therapy alone or in combination with either transdermal or oral testosterone (Table 2).

Leucine rate of appearance

In the absence of GH, neither transdermal nor oral testosterone significantly affected LRa (Fig. 3 and Table 2). GH therapy significantly ($P < 0.01$) increased LRa by 15.2 ± 3.9% when compared with baseline. During GH therapy, the addition of transdermal ($P < 0.01$) and oral testosterone ($P < 0.05$) significantly reduced LRa compared with GH therapy alone. The effect on LRa between transdermal and oral testosterone administration was not significantly different (Fig. 3 and Table 2).

TABLE 2. Treatment effects on testosterone, IGF-I, SHBG levels, and protein turnover measures

	No GH			GH		
		Testosterone			Testosterone	
	Baseline	Td	Oral	Baseline	Td	Oral
T (nmol/liter)	2.8 ± 0.6	19.5 ± 3.4[a]	4.3 ± 0.9	3.5 ± 0.5	19 ± 3.4[a,b]	3.7 ± 0.6
IGF-I (nmol/liter)	15.6 ± 2.1	14.5 ± 1.5	14.3 ± 1.5	40.7 ± 3.9[a]	39 ± 3[a]	47.2 ± 4.7[a,b]
SHBG (nmol/liter)	23 ± 3	19.8 ± 2.6[a]	22.2 ± 3.9	23.6 ± 3.3	21.4 ± 2.8	21.8 ± 3.3
LRa (μmol/min)	151.4 ± 11.9	158.9 ± 16	159.2 ± 13.2	173.6 ± 14[a]	161.6 ± 11.5[b]	158.2 ± 9.7[b]
Lox (μmol/min)	37.6 ± 4.1	34.7 ± 3.7	36.9 ± 5.2	38 ± 4.6	32.7 ± 3.2[a,b]	32.7 ± 3.1[a,b]
Lox (% from LRa)	24.7 ± 1.5	21.9 ± 1.3	22.7 ± 1.9	21.7 ± 1.8[a]	20.1 ± 1.1[a]	20.8 ± 1.6[a]

Data are presented as mean ± SEM. T, Testosterone; Td, transdermal.

[a] $P < 0.05$ *vs.* nil (baseline, no GH).

[b] $P < 0.05$ *vs.* GH therapy (baseline, GH).

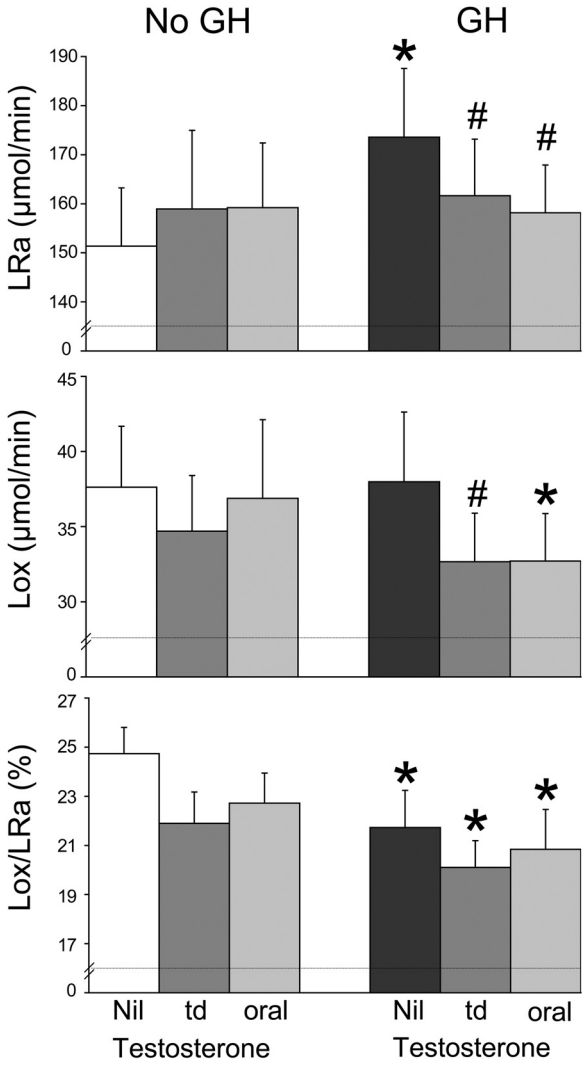

FIG. 3. Effects of testosterone and GH therapy used alone and in combination on whole-body protein turnover. A, LRa, a measure of protein breakdown; B, net Lox, a measure of irreversible loss of protein; C, proportion of leucine oxidized, expressed as percentage from LRa. Data are expressed as means ± SEM. $P < 0.05$: *, *vs.* no treatment; #, *vs.* GH administration. td, Transdermal.

Net Lox

In the absence of GH, when compared with baseline, neither transdermal nor oral testosterone significantly affected Lox (Fig. 3 and Table 2). Lox did not significantly change during GH therapy. During the GH therapy phase, the addition of transdermal testosterone significantly reduced Lox by 11.1 ± 5.2% compared with baseline ($P < 0.05$). There was also a significant reduction of 10.7 ± 4.3% compared with GH therapy alone ($P < 0.05$). During GH therapy, the addition of oral testosterone significantly reduced Lox by 11.3 ± 3.5% compared with baseline. There was also a significant reduction of 10.2 ± 4.7% compared with GH therapy alone ($P < 0.05$; Fig. 3 and Table 2). The effect on Lox was not significantly different between transdermal and oral testosterone administration (Fig. 3 and Table 2).

The proportion of leucine oxidized

We next analyzed the data to determine which intervention induced a significant reduction in leucine oxidized as a proportion of LRa (percent Lox/LRa), because this proportion is an inverse measure of protein synthesis. ANOVA revealed that the percent Lox/LRa was not significantly affected by testosterone treatment alone. This proportion was significantly ($P < 0.05$) reduced by GH treatment and unaffected by cotreatment with either oral or transdermal testosterone (Fig. 3 and Table 2). The mechanisms by which GH and combined GH and testosterone treatment reduced the proportion of leucine oxidized were different. In the case of GH, the reduction occurred secondary to an increase in LRa with no change in Lox. The addition of testosterone prevented the GH-induced increase in LRa while simultaneously reducing Lox, resulting in a net lowering of the proportion of leucine oxidized (Fig. 3 and Table 2).

Thus, GH and testosterone affect different components of leucine turnover, with an effect of testosterone evident only in the presence of GH.

Discussion

We aimed to determine the site of GH and testosterone interaction in the regulation of whole-body protein metabolism. We employed oral and transdermal testosterone administration to deduce whether the interaction occurred primarily in the liver or in extrahepatic tissues. The testosterone regimen increased systemic androgen exposure via the transdermal route or solely hepatic exposure via the oral route (11). In the absence of GH, neither systemic nor hepatic testosterone exposure significantly influenced LRa and Lox. In the presence of GH, both systemic and hepatic testosterone administration reduced the LRa, an index of protein breakdown, and the rate of Lox, an index of irreversible loss of protein. There was no significant difference between transdermal and oral testosterone administration on protein metabolism during GH therapy. In the doses used, neither GH nor testosterone alone reduced Lox, a measure of irreversible loss of protein. Only when testosterone was combined with GH was there a measurable net beneficial anabolic effect.

The liver is a major site of protein metabolism. Its contribution to whole-body protein synthesis approximates that of muscle, whereas its protein oxidation rate is about half that of muscle (18). In the postabsorptive state, the rate of protein degradation exceeds that of synthesis in muscle but not in the liver, indicating that during substrate deprivation, amino acids are supplied to the liver by skeletal muscle (18). Thus, it is a critical site for regulation of whole-body protein metabolism. This study provides

strong evidence that GH and testosterone interact in the liver to regulate whole-body protein metabolism.

Our study shows that GH and testosterone act on different components of protein metabolism. In the postabsorptive state, about 20% of amino acids derived from proteolysis are lost irreversibly through oxidation (Fig. 2). The amount of leucine oxidized is dependent on 1) the LRa, an index of protein breakdown, and 2) the partitioning of leucine to oxidation pathway, representing a proportion of leucine oxidized. GH treatment alone reduced the proportion of leucine oxidized while stimulating the rate of leucine turnover, which resulted in no net change in the amount of leucine oxidized. Testosterone alone did not influence the proportion of leucine oxidized or the rate of leucine turnover. However, testosterone abrogated the stimulation of leucine turnover but did not affect the reduction in proportion of leucine oxidized induced by GH, resulting in a net reduction in the amount of leucine oxidized. Because the reduction in Lox results in a reduction in irreversible loss of protein, this represents a net anabolic effect arising from a partitioning of amino acid utilization toward protein synthesis. Our data show that GH and testosterone act in a complementary way on different components of protein metabolism in exerting net anabolic effect.

We observed that GH enhanced leucine turnover while increasing the fraction that is recycled to protein synthesis after breakdown. We did not observe a significant reduction in Lox by GH therapy alone, as reported in some studies previously (3, 19–21). This apparent discrepancy may reflect a time-dependent effect of GH on the dynamics of protein metabolism. In the early weeks of GH replacement, Lox is reduced together with an increase in whole-body leucine turnover (3, 19–21). After 3 months of GH replacement, an effect on Lox is lost, but the increase in leucine turnover with recycling of leucine for protein synthesis is maintained reflecting a new steady state (3). Because the GH effect on protein metabolism is time dependent, we designed the study so that the effects of testosterone could be studied against a stable baseline of GH therapy.

The design does not allow us to determine whether the site of GH action is extrahepatic, hepatic, or both. Recycling of amino acids may occur locally in muscle and other tissues or occur at a systematic level involving a central role of the liver in recycling of nitrogen to and from peripheral tissues and disposal via the urea cycle. Studies in rodents have reported that GH reduces hepatic nitrogen clearance by inhibiting urea synthesis, which results in an increase in nitrogen content of liver and muscle (22). Urea synthesis and hepatic nitrogen clearance are suppressed by GH administration in healthy men and in GH-deficient patients (23–25). Because urea formation represents an irreversible pathway for nitrogen loss, the data suggest that GH enhances the recycling of nitrogen for reuse in protein synthesis. It is conceivable that the partitioning of leucine toward protein synthesis by GH, as observed from the leucine turnover study, may in part be represented by a reduction in nitrogen clearance in the liver.

TABLE 3. Effects of GH alone and combined with testosterone on whole-body protein metabolism and the proposed sites of interaction

		Protein		Interaction	
	T	Breakdown	Oxidation	Hepatic	Extrahepatic
GH		↑	↔		
GH	Oral	↓	↓	√	—
GH	Td	↓	↓	√	√

GH therapy alone increased protein breakdown. Addition of oral testosterone reduced protein breakdown and oxidation. Because oral testosterone exerts solely hepatic effect, the site of GH and testosterone interaction is liver. Addition of transdermal testosterone also reduced protein breakdown and oxidation. Because transdermal testosterone exposes liver and extrahepatic sites to testosterone, if the interaction site would have been primarily extrahepatic, the effect on protein metabolism would differ between oral and transdermal testosterone administration. However, this was not so. Therefore, the site of GH and testosterone interaction is liver. T, Testosterone; Td, transdermal; ↑, increase; ↓, decrease; ↔, no change; —, no interaction; √, site of GH and testosterone interaction.

We showed that, regardless of the route of administration, in the presence of GH, testosterone reduces leucine turnover (Table 3). It is unlikely that testosterone inhibited the peripheral breakdown of protein induced by GH. Had this been the case, then transdermal and not oral testosterone would have modified the process. However, this was not observed. These data may indicate a dynamic process of proteolysis occurring in the liver under GH stimulation that is attenuated by testosterone. What this means physiologically is unknown. Turnover studies involving the tracking of liver-specific proteins or transhepatic arteriovenous cannulation studies are required to elucidate the significance of the present findings.

Our study indicates that testosterone treatment alone did not affect whole-body protein metabolism. However, we cannot exclude the possibility that testosterone may have exerted a significant effect had the dosage been higher. Testosterone exerts dose-dependent effect on lean body mass (26). We previously showed that in the absence of GH, administration of testosterone by the im route inhibits irreversible loss of protein (9), an observation at odds with the present findings. It is likely that the pharmacological levels of testosterone achieved after im injection imparted a greater effect on protein metabolism. In the present study, the mode and method of testosterone administration resulted in physiological concentrations in

blood (11). Our results also indicate that a whole-body anabolic effect can be achieved by using a physiological dose of testosterone administered via oral route free of any systemic androgenic effects. This finding is of potential therapeutic benefit because systemic testosterone administration is associated with an increased risk of cardiovascular adverse events and androgenic effects in women, both of which can be avoided by oral administration of a dose that imparts physiological hepatic effects (27). Therefore, testosterone administration via oral route may open new treatment strategies for frail elderly. Further research is required to provide a definite conclusion on the therapeutic implications of our findings.

In summary, in the absence of GH, neither transdermal nor oral testosterone exposure significantly influenced LRa and Lox. GH therapy significantly stimulated whole-body LRa, an effect abrogated by the addition of testosterone regardless of the route of administration. Net Lox was reduced by combined administration of GH and testosterone but not by either hormone alone.

We conclude that in the doses used, only in the presence of GH, testosterone stimulates anabolism by reducing protein breakdown and oxidation. Because the net effect on protein metabolism during GH therapy is not different between systemic and solely hepatic testosterone administration, the liver is the primary site of interaction between these two hormones. In the GH-sufficient state, oral testosterone administration of a physiological dose can induce a whole-body anabolic effect without systemic androgenic effects. These findings have potential benefit for treatment of frailty in both men and women.

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