Bone and Body Composition Analyses by Dxa in Adults With Gh Deficiency: Effects of Long-term Replacement Therapy

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Abstract

Purpose: The effects of growth hormone (GH) replacement on bone mass and body composition in adult with GH deficiency (AGHD) are still debated with regard to their persistence in the long term. Moreover, the impact of gender on the response to GH is controversial. Aim of this study was to evaluate the long-term effects of rhGH replacement on bone mass and body composition in a monocentric cohort of patients with AGHD.

Methods: Data from 138 patients with AGHD (34.3 ± 14.3 years, 48 women and 90 men) treated with rhGH for a period of at least 3 years up to a maximum of 10 were retrospectively collected. Bone mineral density (BMD) at the lumbar spine, femur, and radius, and total and truncular body composition were evaluated by dual energy X-ray absorption (DXA) before and during treatment. Clinical and laboratory evaluations were performed before and during the treatment period on an annual basis.

Results: Lumbar spine BMD consistently increased in males, while in females decreased after a transient improvement observed during the first 4 years of therapy. There were no significant changes in femoral BMD in either sexes, while a progressive increase at radius was observed only in males. Lean mass significantly increased in both sexes, while fat mass decreased only in males.

Conclusions: In AGHD patients long-term rhGH replacement therapy induces a positive effect with regard to bone mass and body composition. A sexual dimorphism in the response to treatment is evident, with males displaying more favorable outcome.

Introduction

Untreated growth hormone deficiency (GHD) in adulthood is characterized by decreased bone mineral density (BMD) [1, 2] and body composition abnormalities such as increased fat mass, predominantly in the abdominal (visceral) compartment [3], as well as a reduced lean mass at the expense of the muscles [4, 5]. The clinical outcomes of these features on fracture and cardiovascular risks are debated for the lack of compelling data in the adult population. Nevertheless, it is generally assumed that GHD in adulthood may contribute to the increased fracture risk [6, 7] and the reduction of cardiac functionality observed in patients with panhypopituitarism [8]. In fact, a lower fracture incidence was recorded in GHD patients receiving growth hormone as compared with those who did not, even after adjustment for confounding factors in multivariable analysis [9]. Equally, as extensively discussed [10], GH replacement therapy benefits body composition by enhancing lean mass [11–13] as well as heart function by improving echocardiographic morphology [14–15].

The positive effect of replacement therapy with recombinant human GH (rhGH) on bone mass, body composition and cardiovascular functions has been consistently reported for treatment periods lasting up to fifteen years [16–21]. However, the effects were not constant among genders and over time.

On the hypothesis that GH replacement in adulthood activates different metabolic pathways in the bone of male and female patients, our group specifically evaluated the gender-dimorphic effect on the bone outcome under 3 years of rhGH treatment [22]. The results showed that in both genders GH replacement was able to activate bone remodeling, but bone mass outcome assessed by x-rays-absorptiometry (DXA) was only positive in males, even after controlling for potential confounders. These data were later confirmed in a subsequent study reporting a greater increase in men than in women of the bone mineral density (BMD) at all skeletal locations measured by DXA scans [20]. However, in a previous study from the same institution, women resulted to have a greater benefit than male by rhGH therapy, particularly those on estrogen replacement therapy [23]. Moreover, in a retrospective analysis of data extracted from KIMS (Pfizer International Metabolic Database), an international survey of adult GHD patients from 31 countries treated for 4 years with rhGH, men showed a greater increase in lumbar spine BMD over baseline than women; however, the association was lost on multivariate analysis [24]. Therefore, the observation that the bone responsiveness to GH replacement in adult GHD varies as function of sex remains controversial.

It is also controversial whether or not the bone effect of rhGH lasts over time. Fifteen-year of rhGH replacement in GHD adults engendered indeed a sustained BMD increment in total body and lumbar spine but not in the femur neck, where the BMD peaked at 7 years and then decreased towards baseline [20].

Based on these controversies, we have designed the following study with the aim to retrospectively reassess bone responsiveness to GH replacement in a large population of adult GHD patients followed for up to 10 years, with a specific emphasis to the gender effect and to the maintenance of bone response over time. The re-evaluation of gender effects in the long time is clinically relevant as the interaction among GH, estrogens and androgens replacements might also have an impact on other target body compartments, like fat tissue [16, 17, 20, 22] and the heart [14, 15], functionally connected to bone by a complex and yet not defined interplay.

Materials And Methods

Subjects

Between January 1999 and December 2017, we recruited 169 patients with AGHD treated with rhGH replacement therapy and followed-up in the outpatient clinic of the San Raffaele Scientific Institute in Milan.
Patients with rhGH treatment of less than 3 years were subsequently excluded from the study. The final population included 138 patients (48 females and 90 males, aged 34.3 ± 14.3 years at baseline, mean ± SD).

The diagnosis of AGHD was confirmed by provocative testing with arginine + GH-releasing hormone. The peak GH level to diagnose GH deficiency was adjusted according to the body mass index of the patients [25]. Patients with multiple pituitary hormone deficiency were on stable replacement therapy for the

deficient pituitary axes for at least 6 months before starting GH replacement. The initial rhGH dose was 0.2 mg/day in most patients; dose was progressively adjusted to maintain circulating IGF-1 levels within the reference range for age. To assess the adequacy of rhGH replacement, we conducted a retrospective analysis to evaluate how many patients showed IGF-1 values within the therapeutic goal at the end of the study period. At the last available observation, 110 out of 138 patients (79.7%) achieved the therapeutic goal, while in 8 patients IGF-1 levels were below and in 20 under the reference range, respectively. The percentage of patients within the therapeutic goal was similar in males and females (78,9% vs 81,2%, respectively).

When vitamin D insufficiency and/or elevated parathyroid hormone levels were detected, oral calcium and vitamin D3 supplementation were prescribed.

Data collection was retrospective and did not affect routine clinical care.

Onset of GHD occurred during adulthood in 87 patients (63%) and during childhood in 51 patients (37%). All patients with childhood-onset GHD were treated with rhGH replacement therapy until completion of the linear skeletal growth; in patients with congenital GHD, rhGH was then withdrawn, and arginine-GHRH test was repeated after at least one month to confirm GHD persistence in adult age. Patients with GHD due to an organic disease, such as craniopharyngioma, did not require GH retesting.

In the majority of patients, pituitary deficiency was secondary to a tumor of the pituitary region and/or its treatment, as follows: pituitary adenoma (n = 27 non-functioning adenoma, n = 7 prolactinoma, n = 7 FSH-producing adenoma, n = 2 GH-producing adenoma, n = 2 apoplectic adenoma), craniopharyngioma (n = 39), germinoma (n = 4), parasellar meningioma (n = 3), medullo-blastoma (n = 1), hypothalamic astrocytoma (n = 1), and granular cells tumor (n = 1). In the remaining patients the causes of pituitary deficiency were neurosarcoidosis (n = 2), radio-therapy for orbital rhabdomyosarcoma (n = 2), optic nerve glioma (n = 2), Langerhans cell histiocytosis (n = 3), lymphocytic hypophysitis (n = 2), empty sella syndrome (n = 4), idiopathic panhypopituitarism (n = 9), idiopathic isolated GH deficiency (n = 10), idiopathic combined GH and gonadotropins deficiency (n = 2) or GH and TSH deficiency (n = 2), pituitary hypoplasia (n = 4), Sheehan's syndrome (n = 2), pituitary cyst (n = 2), and Kabuki syndrome (n = 1). Twenty-seven patients had congenital GHD. Sixteen patients had isolated GHD. One hundred twenty-two patients had at least one additional pituitary hormone deficiency, with 45 of these having complete pituitary insufficiency. Fifty-four patients had diabetes insipidus. Among the 48 female participants, ten had a normal gonadal function, 12 were on oral estrogen hormone-replacement therapy (HRT), and 16 were on transdermal HRT; in the remaining 10 women the diagnosis of hypopituitarism was made after the achievement of menopause. All gonadotropin-deficient men (n = 71) received testosterone replacement.

Age, age of GHD onset, BMI, and proportion of individuals with childhood-onset GHD were similar in males and females. Baseline patient characteristics are reported in Table 1.

Table 1
Baseline clinical characteristics of 138 AGHD patients

	Males *(n = 90)*	**Females** *(n = 48)*	**p-value**
Age *(years)*	33.9 ± 15.0	35.1 ± 14.2	0.65
BMI *(Kg/m²)*	26.51 ± 5.52	25.97 ± 4.66	0.58
GHD onset *n (%)*	19 (21.1)	8 (16.8)	0.78
Congenital	16 (17.8)	8 (16.8)	
Childhood	55 (61.1)	32 (66.6)	
Adult age			
Age of GHD onset	30.7 ± 16.7	27.4 ± 17.9	0.31
Concomitant pituitary deficiencies *n (%)*	11 (12.2)	5 (10.4)	0.95
None (isolated GHD)	59 (65.6)	34 (70.8)	0.53
ACTH	70 (77.8)	42 (87.5)	0.16
TSH	71 (78.9)	28 (73,6) [a]	0.52
FSH/LH	32 (35.6)	22 (45.8)	0.24
Diabetes insipidus			
Laboratory results	115.33 ± 81.19	80.28 ± 70.95	**0.01**
IGF-1 *(ng/mL)*	56.15 ± 31.39	57.15 ± 30.41	0.70
PTH *(pg/mL)*	19.25 ± 13.63	20.03 ± 11.86	0.56
25-hydroxyvitamin D *(ng/mL)*			

BMI: Body Mass Index; GHD: Growth hormone deficiency; ACTH: Adrenocorticotrophic Hormone; TSH: Thyroid Stimulating Hormone; FSH: Follicle Stimulating Hormone; LH: Luteinizing Hormone; IGF-1: Insulin-like Growth Factor-1; PTH: Parathyroid Hormone.

[a] Ten out of 48 patients were excluded from the analysis since diagnosis of GHD was made after menopause

Dual-Energy X-Ray Absorptiometry

The areal bone mineral density (BMD) at the lumbar spine vertebrae, radius (one-third proximal), and femur (neck and total) was measured using the Hologic (Waltham, MA) QDR 4500 at 0, 6, and 12 months, and then at variable intervals based on clinical indication. At femur, the assessment of bone area was also included. The coefficients of variation (CVs) for the measurements were calculated daily by quality-control scans of spine phantoms and were < 0.5%. Furthermore, according to the manufacturer's

recommendations, the stability of the densitometer was also documented by yearly measurement of the European spine phantom (ESP) 026.

The reference values provided by the manufacturer were used to calculate T- and Z- scores, which express individual BMD values as SD scores in relation to the normal mean BMD values of a same sex healthy population of a reference age (25–30 yrs) and of a similar age, respectively.
According to the International Society for Clinical Densitometry guidelines [26], we used Z scores in patients under 50 years and T scores in patients over 50 years.

Body composition was also assessed with DXA scanner, measuring total and trunk lean and fat mass. Estimated margin of error for body composition was between 2 and 5%.

The same operator (M. S.) performed all dual-energy X-ray absorptiometric (DXA) measurements using the same equipment.

Laboratory Assays

Measurements of blood biochemical parameters were performed at 0,6,12 months and then annually up to a maximum of ten years.

Serum IGF-1 was measured with a chemiluminescence assay (Immulite 2000; Medical System, Genoa, Italy). The sensitivity of the assay was 20 ng/ml; intra-assay CVs were 2.9% in the lower range and 4.9% in the upper range; interassay CVs were 4.3 and 7.1%, respectively. Plasma total calcium was measured using the Advia 2400 analyzer (Siemens Medical Solutions Diagnostics, Tarrytown, NY). Plasma intact PTH levels and plasma 25(OH)D_3 levels were measured with a chemiluminescence immunoassay (CLIA) on Architect analyzer (Abbott, Green Oaks, Illinois) until 2016, on Roche analyzer (Rotkreutz, CH) afterwards.

Statistical Analysis

Categorical variables are presented as frequencies and percentages, continuous variables as means ± SDs. Comparisons between groups were performed, for categorical variables, using the X^2 test or Fisher's exact test, as appropriate. Continuous variables were compared using the unpaired two-tailed Student T test or the Wilcoxon's rank sum test, if variables were not normally distributed (according to the Shapiro-Wilk statistic).

The BMD, bone area and parameters of body composition were the outcome variables in mixed-effects models with repeated measures over time. Age, age of onset of GHD, pre-treatment body mass index (BMI), and BMD were included in every model. All tests were two-tailed, and *p*-values < 0.05 were considered significant. Statistical analyses were performed using the SAS software version 9.4 (Cary, NC: SAS Institute Inc, USA).

Results

Baseline IGF-1 levels were higher in males than in females ($p = 0.01$; Table 1). GH replacement increased serum IGF-1 levels over time among both males ($p = 0.02$) and females ($p = 0.04$). However, IGF-1 levels over time were higher in males ($p = 0.01$), despite the fact that females were prescribed a higher mean GH dose per kilogram of body weight ($p < 0.01$). Mean serum IGF-1 levels and mean GH daily dose during GH replacement are shown in Fig. 1.

Mean calcium, PTH and vitamin D levels were within the reference ranges both at baseline and during the study period, without differences between males and females (data not shown).

Baseline DXA measurements were available for 126 patients (Table 2). All BMD parameters were higher in males than in females ($p < 0.01$), as expected. The changes over baseline of BMD during GH replacement are shown in Fig. 2. During GH replacement we observed a BMD increase at lumbar spine among males ($p < 0.01$) that was sustained for the entire follow-up, while in females BMD increased slightly in the first four years of rhGH replacement and then decreased significantly ($p = 0.01$). Femoral BMD did not vary significantly in both sexes during the study period ($p = 0.22$ in males; $p = 0.31$ in females), while BMD at radius increased significantly following rhGH replacement only in males ($p = 0.03$; $p = 0.09$ in females). Femoral bone area during GH replacement remain unchanged as compared to baseline in both sexes (data not shown).

Table 2
Baseline DXA parameters of AGHD patients.

	Males	Females	p-value
T-score	12	8	0.82
Number of patients	-1.04 ± 2.41	-1.70 ± 0.96	0.82
Lumbar spine (L1-L4)	-0.50 ± 1.36	-0.78 ± 0.82	0.62
Femur (total)	-0.86 ± 1.01	-1.15 ± 0.58	0.07
Femur (neck)	-1.27 ± 1.21	-0.67 ± 0.62	**0.04**
Distal radius	68	38	**< 0.01**
Z-score	-0.76 ± 1.37	-1.27 ± 1.01	**< 0.01**
Patients	-0.24 ± 1.11	-0.90 ± 0.91	0.11
Lumbar spine (L1-L4)	-0.07 ± 1.45	-0.94 ± 1.04	
Femur (total)	-1.07 ± 1.37	-0.61 ± 0.85	
Femur (neck)			
Distal radius			
BMD (g/cm²)	80	46	**< 0.01**
Patients	0.989 ± 0.172	0.877 ± 0.108	**< 0.01**
Lumbar spine (L1-L4)	0.986 ± 0.167	0.836 ± 0.114	**< 0.01**
Femur (total)	0.889 ± 0.186	0.742 ± 0.130	**< 0.01**
Femur (neck)	0.743 ± 0.067	0.639 ± 0.051	
Distal radius			
Area (cm²)	80	46	**< 0.01**
Patients	40.881 ± 4.103	30.859 ± 4.039	**< 0.01**
Femur (total)	5.566 ± 0.376	4.811 ± 0.416	
Femur (neck)			

BMD: bone mineral density

	Males	Females	p-value
Body composition	75	45	**< 0.01**
Patients	55646.79 ± 10670.3	38968.12 ± 6879.79	**< 0.01**
Lean body mass *(g)*	22055.92 ± 7792.36	26212.81 ± 8422.79	**< 0.01**
Fat body mass *(g)*	26.94 ± 5.01	38.44 ± 4.67	**< 0.01**
Fat body mass *(%)*	27219.25 ± 5604.62	19752.56 ± 3727.9	0.18
Trunk lean mass *(g)*	11570.1 ± 4514.32	12694.41 ± 4143.55	**< 0.01**
Trunk fat mass *(g)*	28.48 ± 6.13	37.9 ± 4.92	
Trunk fat mass *(%)*			
BMD: bone mineral density			

Baseline body composition parameters were available for 120 patients (Table 2). Lean body mass and trunk lean mass were higher in males ($p < 0.01$), whereas fat body mass was higher in females ($p < 0.01$).

Following rhGH replacement, total lean mass increased in both sexes ($p < 0.01$ in males; $p = 0.01$ in females) while total fat mass decreased significantly only in males ($p = 0.04$) and remained unchanged in females ($p = 0.43$; Fig. 3). At trunk, we observed a slight albeit significant decrease in fat mass in males ($p = 0.04$) but not in females ($p = 0.06$). Trunk lean mass increased in both sexes ($p < 0.01$ in males; $p = 0.01$ in females), but the extent of the increase was significantly higher in males than in females ($p < 0.01$; Fig. 3).

Discussion

Our study evaluated the effects of long-term replacement with rhGH on a population of adult GHD patients with respect to bone density and body composition, assessed by dual energy X-ray absorptiometry (DEXA).

The DEXA analysis showed that vertebral bone density increased in males and decreased in females in the long term, despite a transient BMD gain in the latter during the first years of treatment. No significant changes were detected in both sexes at femoral level, but females showed a trend towards reduction of femoral neck BMD. BMD increased in males at radius and remained stable in females. Regarding body composition, ten years of rhGH replacement induced a reduction of fat mass and an increase in lean mass. However, the effect on lean tissue was documented in both sexes, while fat mass decreased only in males.

The present results support and extend our previous study [22]. In fact, the long-term observation confirms the existence of a gender specificity in the bone response to rhGH replacement, with males displaying a greater BMD achievement that is sustained over time. The evidence that the bone response

to rhGH varies as a function of sex, being more favorable in males, was not consistent in all studies. Some authors [16, 20] reported a larger BMD increase in males following long-term GH replacement, while others [23] showed similar or even greater benefits in response to rhGH in women as compared to men. Moreover, Tritos [24] reported a positive association between male sex and bone outcomes, that however did not persist on multivariate analysis. In this still unresolved scenario, our observations support the higher sensitivity of the male skeleton to rhGH replacement and its persistence in the long term.

The mechanisms underlying this gender specificity remain matter of speculation. Growth hormone exerts its effect on bone both directly and through stimulation of IGF-1 production. Since IGF-1 hepatic generation in response to rhGH is lower in females than in males, due to the inhibitory action of circulating estrogens on liver function [27], it has been hypothesized that women are less responsive to rhGH replacement due to lower circulating IGF-1 [24]. According to this view, our study showed that IGF-1 levels were higher in males than in females. However, the percentage of patients reaching their therapeutic goal was similar in males and females, thus excluding the possibility that the different bone response observed in females was influenced by undertreatment. Furthermore, in our previous study [22] we excluded a significant correlation between circulating IGF-1 and bone outcomes. In this regard, it has to be considered that rhGH exerts its osteoanabolic action mainly through increased bone expression of IGF-1 [28], which is not necessarily reflected by circulating IGF-1 levels.

Another factor that has to be taken into account is the direct effect of GH on bone. Growth hormone modulates the crosstalk between osteoblasts and osteoclasts [29, 30], influencing the balance between bone apposition and resorption. High rhGH doses may excessively stimulate bone resorption and limit its favorable effects on the skeleton, at least during the early years of treatment [31]. Since in our study females were prescribed higher rhGH doses than males, this could explain, at least in part, the different bone response observed between sexes.

Besides the direct effects of GH and IGF-1 on bone cells, we can speculate that several bone regulatory metabolic pathways, with a gender related outcome, might be activated by rhGH replacement. Conditions of relative GH excess in comparison to IGF-1 may inhibit osteoblast activity *via* increased sclerostin levels. Uncontrolled acromegaly is indeed characterized by a nonlinear relationship between GH and IGF-1 due to saturation of GH receptors [32], and uncontrolled acromegalic females displayed higher sclerostin levels as compared to both male patients and healthy controls [33]. This may also have occurred in our female population, characterized by higher rhGH doses and lower IGF-1 levels as compared to males.

The interaction between bone metabolism and changes in body composition could also contribute to the gender difference in bone response to rhGH. The increase in lean mass may mediate the osteo-anabolic action of GH, since muscles possess both a biomechanical and a biochemical action, through a paracrine regulation of several myokines and growth factors including IGF-1 [34, 35]. In particular, the increase in muscle mass is associated with a reduction of the synthesis of myostatin [35], which inhibits osteoblast action and promotes differentiation of cells of the osteoclastic lineage [35]. Moreover, it has been reported that rhGH replacement increases the circulating levels of irisin [36], a myokine able to increase

bone cortical mass by stimulating the osteoblast pathways [37], in close relationship with changes in body composition [36]. It can thus be speculated that the more favorable effect on lean mass that we observed in males after rhGH replacement could also have induced a greater bone response.

It has to be taken into account that the existence of a gender specificity in the responsiveness to rhGH replacement with regard to body composition is still a matter of debate [11, 13, 38, 39]; our results are in line with those of Franco et al [11] and Mukherjee et al [13], suggesting that males may experience greater changes in fat mass than females in response to GH. The reason for this difference still needs to be elucidated, but some evidences suggest that androgens may play a major role. Testosterone increases tissue responsiveness to GH through enhanced GHR expression [40], thus potentiating its biological action; moreover, androgens and GH exert additive effects in stimulating protein synthesis, increasing resting energy expenditure and promoting fat oxidation [41], and androgen and GH replacement are both necessary to achieve the full anabolic effect in men with hypopituitarism [41]. On the other hand, the impact of estrogens in this setting seems to be less relevant, as suggested by Franco et al [11], reporting a greater improvement in body composition in response to rhGH in man than in post-menopausal women, characterized by very low circulating estrogens.

Our previous hypothesis [22] that in women the presence of estrogen and the GH-mediated reduction of fat mass could influence the balance between peripheral and central effects of leptin in favor of the latter, enhancing its catabolic action on bone, is not confirmed by the present study; indeed, rhGH replacement reduced fat mass in males but not in females, and the bone anabolic action of leptin is mainly mediated by its circulating levels [42], directly correlating with fat mass.

We did not observe significant variations of femoral bone area following rhGH treatment, in line with our previous work [22]. However, we conducted a preliminary evaluation of the geometrical features of proximal femur with HSA (Hip Structural Analysis), which showed a slight but significant increase in the sub-periosteal diameter of the diaphyseal portion of the femur ($p = 0.03$) after two years of treatment, not evident in the other femoral sections (data not shown). Unlike femoral neck and intertrochanteric area, femoral diaphysis is composed only by cortical bone [43], and GH exerts its action with a more pronounced effect on cortical bone [28, 44, 45]. It is therefore possible that rhGH replacement induces dimensional changes by activating bone modeling only in specific bone sites and with a limited extent.

Bone area was similar in males and females at all measured sites, both at baseline and after rhGH replacement, thus excluding the hypothesis that the different bone response observed in the two genders was attributable to differences in the activation of bone modeling at periosteal surface.

Since GH acts more on cortical than on trabecular bone [28, 44, 45], it would be expected a greater BMD improvement at femur and proximal radius, predominantly constituted of cortical bone, than at lumbar spine, composed mainly of trabecular bone. However, data from our male population show the opposite, in agreement with several previous studies [16, 20]. Trabecular bone in the lumbar spine is sensitive to sex steroids, while BMD at femur decrease with age in both sexes [20]. It is therefore possible that the

different response to rhGH observed at various skeleton sites reflects the interaction between GH and sex steroids.

In our study rhGH therapy did not determine any significant change in BMI in both sexes, in contrast to previous studies [21, 46] reporting an increase of BMI after at least ten years of rhGH treatment. This discrepancy may be related, at least in part, to the lower mean age of the population included in our study as compared to that of previous reports.

Limitations of our work relate to its retrospective nature and to the lack of a control group. In this regard, it has to be considered that a long-term placebo-controlled study would be deemed unethical, given the beneficial effect of rhGH replacement in patients affected by GHD. Some GHD patients referring to our institution spontaneously refused rhGH treatment, but their number was too small to represent an adequate control group. Another factor that could have hampered our evaluation is the limited female sample size, that prevented us from stratifying female patients based on gonadal status. On the other hand, strong points of the study lie in the long observational time and in the fact that all patients were followed in the same institution with a standardized management protocol, samples were analyzed in the same laboratory with the same assays, and DXA scans were performed by the same operator with a standardized procedure.

In conclusion, our findings support the existence of a gender difference in the response to long-term rhGH replacement, with males displaying greater benefits with regard to both body composition and bone mass and retaining them over time. Females showed only a transient and site-specific effect on bone, and a limited positive effect on lean but not fat mass. Potential paracrine and endocrine crosstalks with effect on bone, activated by rhGH replacement and regulated by the concerted action of GH and gonadal steroids, may explain this gender specificity and deserve further investigations.

Declarations

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Avalaibility of data and material: Data are available on request from the corresponding author.

Code availability: Not applicable

Ethics approval: This is an observational study. The San Raffaele Scientific Institute Research Ethics Committee has confirmed that no ethical approval is required.

Consent to participate and consent for publication: Verbal informed consent was obtained from all individual participants included in the study.

Authors contribution: All authors contributed to the study conception and design. Material preparation and data collection were performed by Roberto Lanzi, Francesca Perticone, Marcella Sirtori, and Marco Losa. Statistical analysis was performed by Carlotta Galeone, Claudio Pelucchi, and Mario Pennacchioni. The first draft of the manuscript was written by Alessandro Rossini and Alessandro Rubinacci, and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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Figures

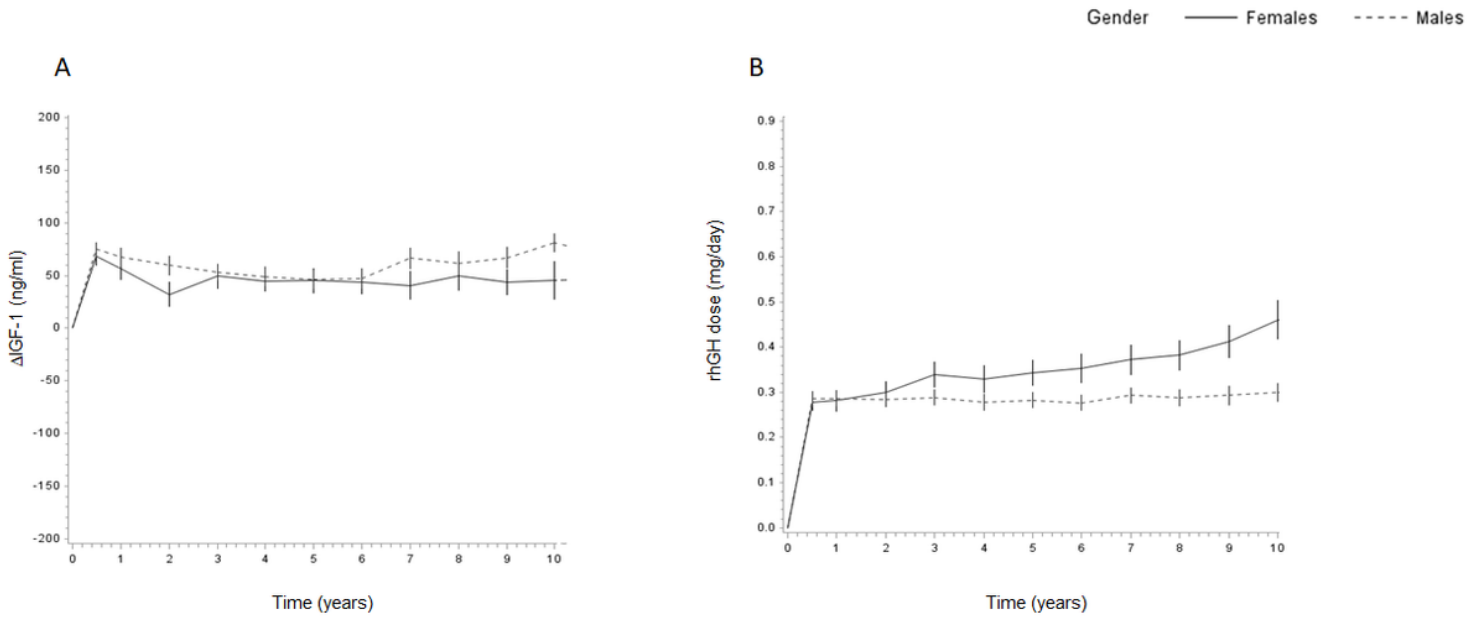

Figure 1

Mean serum IGF-1 levels (panel A) and mean rhGH daily dose (panel B) of AGHD patients during study period. IGF-1 levels are expressed as difference from baseline. Solid lines: Females. Dotted lines: Males. Panel A: p=0.02 vs baseline (males); p=0.04 vs baseline (females); p=0.01 females vs males. Panel B: p<0.01 females vs males

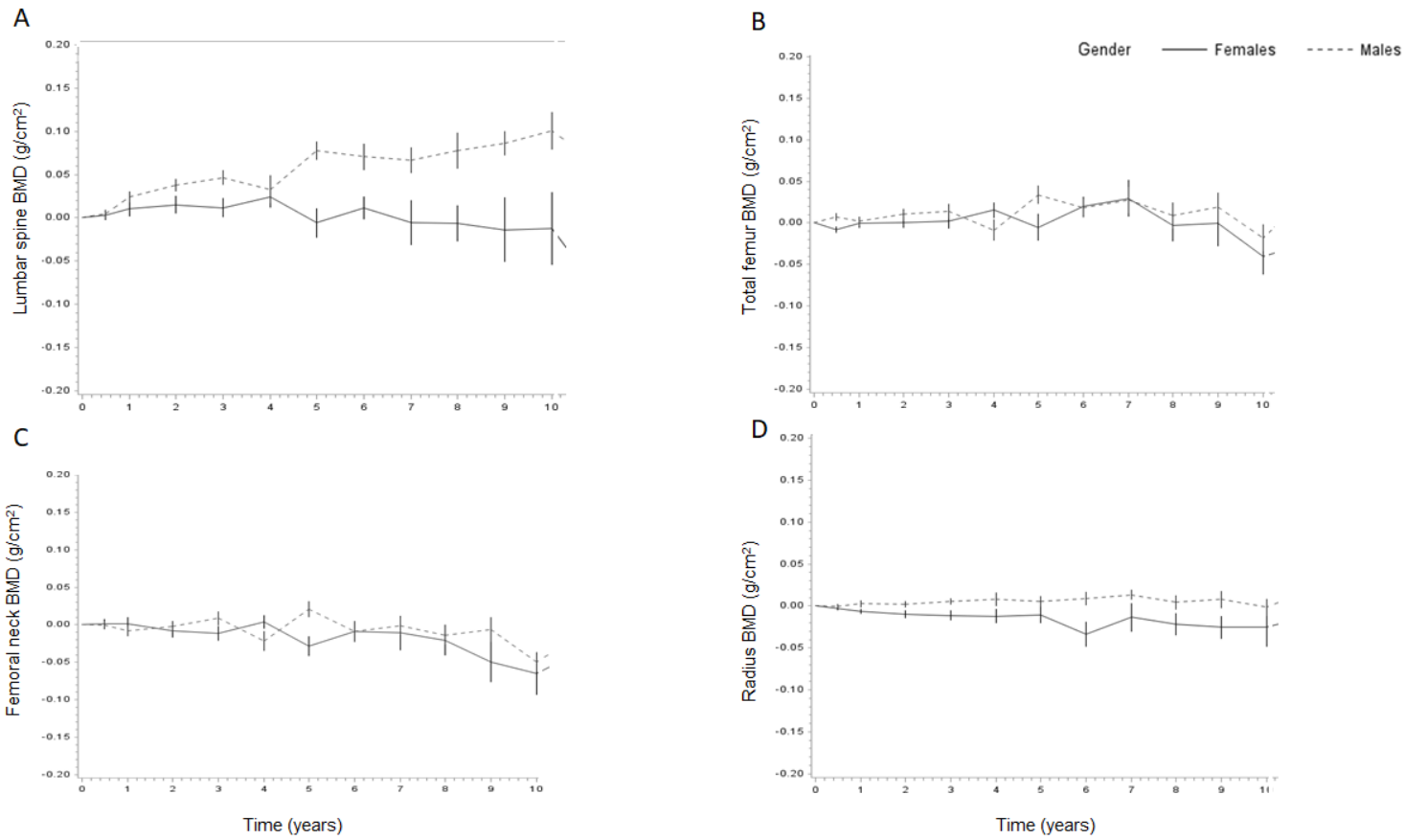

Figure 2

Mean BMD values of AGHD patients during study period. Data are expressed as differences from baseline. Solid lines: Females. Dotted lines: Males. Panel A: Lumbar spine; p<0.01 vs baseline (males); p=0.01 vs baseline (females). Panel B: Total femur. Panel C: Femural neck. Panel D: Ultradistal radius; p=0.03 vs baseline (males)

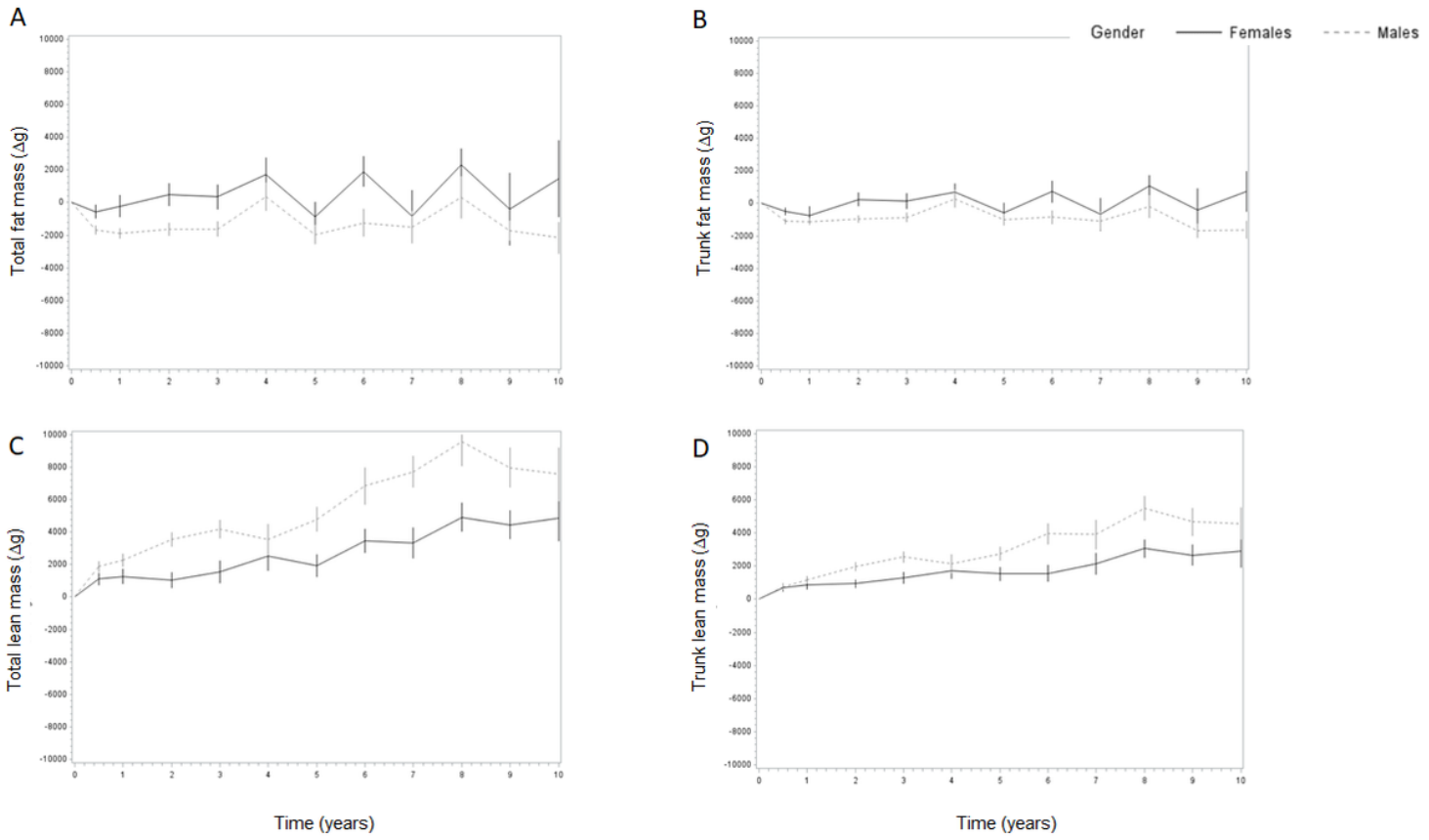

Figure 3

Mean body composition parameters of AGHD patients during study period. Data are expressed as differences from baseline. Solid lines: Females. Dotted lines: Males. Panel A: Total body fat mass; p=0.04 vs baseline (males) Panel B: Trunk fat mass; p=0.04 vs baseline (males) Panel C: Total body lean mass; p<0.01 vs baseline (males); p=0.01 vs baseline (females) Panel D: Trunk lean mass; p<0.01 vs baseline (males); p=0.01 vs baseline (females); p<0.01 females vs males