CLINICAL STUDY

Effects on insulin sensitivity and body composition of combination therapy with GH and IGF1 in GH deficient adults with type 2 diabetes

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Abstract

Objective: The aim of this trial was to evaluate the effect on insulin sensitivity and body composition of combination therapy with GH and IGF1 in adults with GH deficiency (GHD) and diabetes.

Design, patients and methods: A 6-month randomised placebo-controlled pilot study. Fourteen adults with GHD and type 2 diabetes were included. All received rhGH (0.15 mg/day for 1 month and 0.3 mg/day for 5 months) and were randomised to rhIGF1 (15 µg/kg per day for 1 month and 30 µg/kg per day for 5 months) or placebo. Insulin sensitivity was evaluated with euglycaemic hyperinsulinaemic clamp and body composition by computed tomography of abdominal and thigh fat, as well as bioimpedance.

Results: Twelve patients completed the study. They were overweight and obese; at baseline, insulin sensitivity (*M*-value) was low. IGF1 and IGF1 SDS increased in both groups, with the highest increase in the GH and IGF1 group. Positive changes in *M*-value by +1.4 mg/kg per min, in subcutaneous abdominal fat by –60.5 ml and in fat-free mass by +4.4% were seen in the GH and IGF1 group. Corresponding values in the GH and placebo-treated group were –1.5 mg/kg per min, +23 ml and –0.04% respectively ($P=0.02$, $P=0.04$ and $P=0.03$ for delta values between groups). No safety issues occurred.

Conclusions: Combined GH and IGF1 treatment resulted in positive, but rather small effects, and might be a treatment option in a few selected patients.

European Journal of Endocrinology 167 697–703

Introduction

GH exerts insulin-antagonistic effects on glucose and lipid metabolism, as it increases hepatic glucose production, decreases peripheral glucose usage and increases lipolysis, but has anabolic effects on protein metabolism (1). GH regulates the production of insulin-like growth factor 1 (IGF1), which suppresses hepatic glucose production and stimulates peripheral glucose uptake, but like GH, IGF1 has anabolic effects on protein metabolism (1). GH-deficient adults are characterised by insulin resistance, central obesity, dyslipidemia and an increased risk of cardiovascular morbidity and mortality (2, 3, 4). GH replacement improves lipid and body composition parameters, while the metabolic defects affecting the insulin resistance persist (4, 5), which may be related to the GH-induced lipolysis and to hyperinsulinaemia (1).

Recombinant human IGF1 therapy is approved for the long-term treatment of growth failure in children

with primary IGF1 deficiency (6). IGF1 treatment in adults has been evaluated in a few studies. In postmenopausal women treated with IGF1 alone, glucose tolerance improved (7) and in insulin-resistant individuals insulin sensitivity increased (8). IGF1 in combination with GH in healthy volunteers and patients with traumatic brain injury enhanced GH's anabolic effects (9, 10) and in post-menopausal women led to fat mass reduction (11).

GH is a registered treatment in adults with severe GH deficiency (GHD), but it has sometimes been avoided or discontinued in GH-deficient adults with impaired glucose tolerance or diabetes because of GH's anti-insulinemic effects. A potential treatment in these specific patients could be a combination of GH and IGF1 (12) in order to achieve the anabolic effects of both medications as well as the relative improvement in glucose metabolism induced by IGF1.

The aim of this study was to evaluate the effect on insulin sensitivity and body composition of combination

therapy with GH and IGF1 in adults with GHD and type 2 diabetes. The hypothesis was that the combination therapy would improve insulin sensitivity and increase the effect of GH on body composition.

Materials and methods

Study protocol

The study was a 6-month randomised placebo-controlled trial. All participants received rhGH treatment (NutropinAq; 0.15 mg/day for 1 month and 0.3 mg/day for 5 months) injected s.c. every evening and were randomised to rhIGF1 (Increlex) 15 µg/kg per day for 1 month and 30 µg/kg per day for 5 months or placebo (injected s.c. once daily). Doses were based on our clinical experience and on the previous publications. Because of the double-blind design, doses were fixed in both groups and IGF1 levels were not known to the investigators.

Only adults with verified GHD and non-insulin treated type 2 diabetes were included. Patients already on GH replacement therapy discontinued GH treatment 3 weeks before study entry.

All had received relevant instructions in dietary and lifestyle intervention measures in accordance with standard guidelines. Specific oral anti-diabetic medications (glucose-lowering therapy, anti-hypertensive and lipid-lowering therapy) were unchanged throughout the study period.

At baseline, 1, 3 and 6 months anthropometry and investigations were carried out in the morning with patients in the fasting condition. Full examination of the participants was performed at baseline and 6 months. Blood samples were analysed for *P*-glucose, HbA1c, blood lipids, IGF1, IGF-binding protein 1 (IGFBP1), S-total adiponectin, liver transaminases, creatinin and

S-amylase. IGF1 was monitored by an independent expert.

The regional committee for medical ethics in Stockholm (Karolinska Institute) approved the study, and all patients gave their consent for study participation before sample collection and examinations.

Patients

The diagnosis of GHD combined with type 2 diabetes is rare. Nine men and five women with known pituitary diseases of different diagnosis confirmed GHD according to accepted guidelines and with type 2 diabetes were included (Table 1). All had adult onset GHD. Three had isolated GHD, all with relevant pituitary pathology. Eleven were treated with GH and 11 were on stable replacement for other pituitary insufficiencies. Thirteen were treated with oral anti-diabetics, one with lifestyle intervention, 11 received medical treatment for hypertension and four patients were treated for hypercholesterolaemia. All medical therapy was stable for more than 1 year.

Anthropometry and body composition

Physical examinations included measurements of height, weight and waist circumference. Total body fat (kg), body fat percent (%), fat-free mass (FFM; kg) and total body water (TBW; kg) were measured using a body composition analyser, Tanita TBF-300. The measurements were adjusted to age, gender and weight. Bioimpedance is a safe and non-invasive technique for the assessment of human body composition and it measures the resistance of body tissues to a low electrical signal. The principle of bioimpedance is passing an electrical current through the body at different rates depending on body composition and thereby estimating body fat and FFM.

Table 1 Baseline characteristics of 14 patients with GHD and type 2 diabetes assigned to treatment with GH and IGF1 or GH and placebo.

Treatment group	Age (years)	Gender	Pituitary disease	Pituitary insufficiency	Diabetes treatment
GH and IGF1	52	Male	Craniopharyngeoma	FSH/LH, TSH, ACTH, GH	Glibenclamide
	55	Male	Empty sella	FSH/LH, GH	Metformin
	57	Male	TBI	FSH/LH, GH	Metformin
	65	Male	Prolactinoma	GH	Metformin
	56	Female	Prolactinoma	GH	Metformin
	48	Male	NFPA	GH	Metformin
	61	Female	Cushing's disease	FSH/LH, TSH, GH	Metformin
	36	Female	Irradiation of skull in childhood	FSH/LH, TSH, GH	Metformin
GH and placebo	64	Male	NFPA	FSH/LH, TSH, ACTH, GH	Metformin
	53	Female	Idiopathic	TSH, GH	Metformin
	63	Male	Prolactinoma	FSH/LH, TSH, ACTH, GH	Metformin
	48	Male	Craniopharyngeoma	FSH/LH, TSH, GH	Lifestyle Intervention
	69	Female	Idiopathic	FSH/LH, ACTH, GH	Glibenclamide
	64	Female	NFPA	TSH, GH	Metformin DPP4-inhibitor

IGF, insulin-like growth factor 1; TBI, traumatic brain injury; NFPA, non-functioning pituitary adenoma.

Examination of abdominal fat mass was performed with computed tomography (CT). The patients were scanned at baseline and 6 months, using a 16-slice scanner (GE LightSpeed Pro 16). The basic thin slices were combined for volume calculations. Abdominal scans were made at L2–L3 level, and total abdominal fat volume (TF) and visceral abdominal fat volume (VF) were measured. Subcutaneous abdominal fat volume (SF) was calculated by subtracting VF from TF. Measurements of thigh fat volumes were made midway between the right greater trochanter and the joint facet of the lateral condyle.

Assays

HbA1c was measured by cation exchange chromatography (MonoS column) with HPLC (Bio-Rad). Plasma triglyceride (TG), total cholesterol, HDL-C and LDL-C were analysed with routine clinical chemistry techniques. S-total adiponectin was measured by RIA kit (Millipore Corporation, Billerica, MA, USA; Linco Research, Inc., St. Charles, MO, USA). IGF1 and IGFBP1 were determined in serum by in-house RIAs (13, 14). Age-related IGF1 SDS were calculated from the regression line of values for 448 healthy adult subjects (15). Measurements for safety included routine analysis of P-glucose, ASAT, ALAT, S-creatinine and S-amylase.

Euglycaemic hyperinsulinaemic clamp (clamp)

Clamp was performed according to De Fronzo *et al.* (16). Intravenous catheters were inserted into the right arm for insulin/glucose infusion. Insulin Actrapid (40 mU/m² per min; Novo Nordisk A/S, Copenhagen, Denmark) was infused along with 20% glucose (Fresenius Kabi, Stockholm, Sweden) during 120 min. The rate of glucose infusion was adjusted to achieve and maintain a blood glucose level of 5.0 mmol/l. Whole-body insulin sensitivity (M-value) was calculated from the amount of glucose infused during the last 30 min of the clamp divided by body weight (kg) and period (min) and expressed as milligram/kilogram per minute.

Compliance

Empty ampoules collected and returned by the patients were calculated as an indicator for compliance.

Statistical analysis

Results are presented as median and range or mean $\pm$ S.E.M. Mann–Whitney *U* test or unpaired *t*-test was used for statistical calculations. Statistical analysis was

Table 2 Baseline characteristics (median and range) in 14 adults with GHD and diabetes type 2 randomised to treatment with GH and IGF1 or GH and placebo.

	GH and IGF1 (n=8)	GH and placebo (n=6)
Age (years)	55.5 (36, 65)	63.5 (48, 69)
Gender (male/female)	5/3	4/2
IGF1 (μ g/l)	111 (77, 24.2)	114 (30, 206)
IGF1 SDS	−1.2 (−2.6, −1.1)	−0.8 (−5.3, −0.8)
BMI (kg/m ²)	35.6 (27.9, 47.2)	31.9 (25.6, 45.8)
Waist (cm)	116 (101, 138)	112 (98, 113)
Blood pressure	138/88 (112/81, 144/96)	137/84 (115/70, 160/100)
F-glucose (mmol/l)	7.2 (5.2, 9.7)	6.6 (4.8, 8.1)
HbA1c (%)	5.7 (5.2, 7.1)	6.0 (5.2, 6.5)
Total cholesterol (mmol/l)	5.3 (3.6, 6.3)	4.9 (3.3, 5.5)
HDL-C (mmol/l)	0.9 (0.8, 1.4)	0.9 (0.4, 1.0)
LDL-C (mmol/l)	2.7 (1.6, 4.2)	3.2 (2.0, 4.3)
Triglycerides (mmol/l)	1.75 (0.9, 6.3)	1.75 (1.0, 2.4)
Adiponectin (mg/l)	4.5 (2.7, 7.2)	3.6 (2.8, 4.9)
IGFBP1 (μ g/l)	21 (7, 49)	25 (13, 60)
Total body water (kg)	48.9 (40.0–56.4)	46.8 (33.0–58.9)
Fat mass (kg) ^a	35.1 (23.8, 63.9)	29.7 (24.8, 84.2)
Fat-free mass (kg) ^a	66.8 (54.7, 77.1)	66.2 (45.1, 80.4)
Total abdominal fat (ml) ^b	1489.5 (1194.9, 2623.8)	1356.0 (1011.3, 2294.7)
Visceral fat (ml) ^b	588.1 (330.5, 887.9)	664.1 (349.0, 743.8)
Subcutaneous fat (ml) ^b	910.2 (654.5, 1980.9)	744.3 (608.2, 1580.9)
Thigh fat (ml) ^b	406.1 (176.0, 1446.9)	278.0 (56.8, 1123.7)
M-value (mg/kg per min) ^c	2.6 (0.5, 4.5)	5.0 (0.5, 6.0)

IGF1, insulin-like growth factor 1; F-glucose; fasting glucose; IGFBP1, IGF-binding protein 1. *P* values for all parameters correspond to testing a null hypothesis of no differences between the two treatment groups using the Mann–Whitney *U* test. No statistical differences were seen between the groups.

^aAs evaluated with bioimpedance.

^bAs measured with computed tomography.

^cAs calculated from euglycaemic hyperinsulinaemic clamp.

Figure 1 Increase in insulin growth factor 1 (IGF1) levels shown as SDS – during the 6-month treatment with GH+placebo ($n=6$) or GH+IGF1 ($n=6$). Data shown as mean + s.e.m. IGF1 increased in both groups ($P<0.05$).

performed using SAS version 9.3 for Windows (SAS Institute Inc., Cary, NC, USA). Statistical analyses of treatment effects were performed in completers only. Statistical significance was set at $P<0.05$.

Results

At baseline

Eight patients were randomised to treatment with GH and IGF1 and six to GH and placebo. Baseline characteristics are presented in Table 1. All were on relevant and stable replacement for the pituitary insufficiencies. Only the youngest woman was on oral oestrogen replacement; all other women were without replacement. Two patients in the GH and IGF1 group and one patient in the GH and placebo group had not previously been treated with GH. Insulin sensitivity was low as calculated from clamp (median M -value in the GH and IGF1-treated group 2.6 mg/kg per min, $n=4$ and in the GH and placebo-treated group 5.0 mg/kg per min,

$n=5$; NS between groups). Normal insulin sensitivity was set as M -values above 8 mg/kg per min according to local instructions. The clamp was not performed in two women in the GH- and IGF1-treated group and was only performed at baseline in the two men, who withdrew. In one woman it was not possible to get intravenous access; in another very insulin-resistant woman euglycaemia was not obtained. In the GH and placebo-treated group one man had erroneously taken glibenclamide before the clamp at 6 months and therefore results were not included. No differences in study parameters between the groups were seen (Table 2).

At 1, 3 and 6 months

IGF1 and IGF1 SDS increased in both groups, with the highest increment at 6 months in the group treated with both GH and IGF1 (median 276 $\mu\text{g/l}$ and 4.8 SDS) compared with the group treated with GH and placebo (median 121 $\mu\text{g/l}$ and 2.5 SDS) ($P<0.001$ both within and between groups) (Fig. 1). At 6 months all patients in the GH and IGF1 group but only one patient in the GH and placebo group had IGF1 SDS values above 2.0. No changes in BMI, waist circumference, blood pressure, pulse, fasting glucose, HbA1c, TC, LDL-C and HDL-C, TG, total adiponectin or IGFBP1 within or between groups were recorded during the study (Table 3; data for BMI, waist circumference, blood pressure, pulse, TC, LDL-C and HDL-C and TG not shown).

Comparison between baseline and 6 months

No changes were seen in total body fat or TBW. Percent FFM increased by 4.4% (NS) in the GH and IGF1-treated group compared with the GH and placebo-treated group (-0.4% (NS); $P=0.03$ for delta values between groups; Fig. 2). Examinations with CT showed no changes in fat mass in thigh, TF or VF, while a decrease in SF by -61 ml (NS) was seen in the GH and IGF1-treated group compared with an increase of $+23$ ml (NS) in the GH

Table 3 Results at baseline, 3 and 6 months (median and range) for fasting glucose, HbA1c, IGFBP1 and adiponectin in adults with GHD and diabetes type 2 randomised to treatment with GH and IGF1 or GH and placebo.

	Baseline	3 Months	6 Months
GH and IGF1			
n	8	8	6
Glucose (mmol/l)	7.2 (5.2, 9.7)	7.1 (5.2, 13.3)	7.1 (5.3, 12.0)
HbA1c (%)	5.7 (5.2, 7.1)	5.8 (5.2, 8.7)	5.6 (5.2, 8.8)
IGFBP1	21 (7, 49)	16 (8, 99)	17 (6, 45)
Adiponectin (mg/l)	4.5 (2.7, 7.2)	3.4 (2.5, 4.9)	3.1 (2.6, 9.6)
GH and placebo			
n	6	6	6
Glucose (mmol/l)	6.6 (4.8, 8.1)	6.9 (5.1, 11.1)	7.1 (5.2, 10.5)
HbA1c (%)	6.6 (5.2, 5.6)	6.3 (5.2, 6.7)	6.6 (5.4, 8.0)
IGFBP1	29 (9, 107)	25 (13, 60)	31 (10, 79)
Adiponectin (mg/l)	3.6 (2.8, 4.9)	2.9 (1.8, 3.5)	3.3 (2.4, 8.0)

P values for all parameters correspond to testing a null hypothesis of no differences between the two treatment groups using the Mann–Whitney U test. No statistical differences were seen between the groups.

Figure 2 Difference in percent fat-free mass (FFM) between baseline and 6 months $\pm$ S.E.M. in adults with GHD and type 2 diabetes randomised to treatment with GH and IGF1 ($n=6$) or GH and placebo ($n=6$; $P=0.03$ for delta values between groups) (upper panel). Differences in subcutaneous abdominal fat between baseline and 6 months $\pm$ S.E.M. in adults with GHD and type 2 diabetes randomised to treatment with GH and IGF1 ($n=6$) or GH and placebo ($n=6$; $P=0.04$ for delta values between groups) (middle panel). Differences in M -values calculated from euglycaemic hyperinsulinaemic clamp between baseline and 6 months in adults with GHD and type 2 diabetes randomised to treatment with GH and IGF1 ($n=4$) or GH and placebo ($n=5$; $P=0.02$ for delta values between groups) (lower panel). * $P<0.05$.

and placebo group ($P=0.04$ for delta values between groups; Fig. 2). A positive effect on insulin sensitivity as evaluated from the M -value calculated from the clamp was seen in the GH and IGF1 group by 1.4 mg/kg per min (NS), while a negative effect was seen in the GH and placebo-treated group by -1.5 mg/kg per min (NS; $P=0.02$ for delta values between groups; Fig. 2).

Safety adverse effects and compliance

No changes in safety parameters were seen (data not shown). In the GH and IGF1-treated group, two men dropped out at 3 months because of fatigue and alcohol abuse respectively. One man was treated with penicillin due to tonsillitis. Otherwise no adverse events occurred. In particular none of the patients in the group treated with GH and IGF1 developed hypoglycaemia and parotid tenderness. For the patients who completed the study, compliance was excellent as judged by the increase in IGF1 and the study medication was taken in accordance with the study protocol.

Discussion

This is the first randomised placebo-controlled trial on the effect of the combined treatment with GH and IGF1 in adults with GHD and type 2 diabetes. Numbers are small, yet the results are unambiguous. Positive effects were seen on insulin sensitivity, percent FFM and SF in the group treated with combined GH and IGF1.

Adherence to the protocol was evidenced by the increase in IGF1 in both arms, of course most pronounced in the group treated with both GH and IGF1. The study medication was well tolerated and no safety issues occurred.

In a previous study, eight GHD patients received 7 days of treatment with GH once daily, continuous infusion of IGF1 or both hormones together. The study showed that addition of IGF1 reversed GH's effect on insulin sensitivity as measured by clamp (17). Our results from the clamp in the patients treated with GH and IGF1 once daily during 6 months were in accordance with these results, although our patients had type 2 diabetes and were much more insulin resistant. The definition of a normal M -value is not established and in a review the mean M -value in non-obese normal glucose-tolerant subjects was found to vary between 4.7 and 8.7 mg/kg per min (18). We defined normal sensitivity as M -value >8 mg/kg per min which might then be considered somewhat high, and potentially decrease the number of GH-deficient patients obtaining a normal insulin sensitivity with treatment with GH and IGF1. Anyhow, we were interested in the differences induced by the combined treatment and in this respect the definition has no significance. As visceral fat did not change, it is our interpretation that the observed changes in M -value between the groups were caused by an improvement in the GH-induced insulin resistance by a direct effect of IGF1 on glucose uptake in the GH and IGF1-treated group. Supportive of this is also that no systematic changes in HbA1c were observed thus not indicating any major changes in the glycaemic control. GH treatment in fixed doses was given to all study participants. GH is known to impair insulin sensitivity

in a shorter perspective of time (1, 4), while a potential improvement in body composition in a longer-term perspective could be expected to improve the insulin sensitivity. Our study had a duration of 6 months and during that time no major improvement of body composition was observed. Different results during longer trials cannot be excluded.

IGFBP1 is considered a short-term regulator of IGF1 bioactivity and insulin is the main inhibitor of hepatic production of IGFBP1. Insulin increases free IGF1 levels and IGFBP1 is positively associated with insulin sensitivity (19). We did not observe any changes in the low-IGFBP1 values indicating that no major alterations in hepatic insulin resistance were induced. Adiponectin levels are known to be low in obesity and type 2 diabetes (20). In this study adiponectin was below the normal range at baseline and remained low during the study. As body composition was largely unchanged, improvements in IGFBP1 and adiponectin were not expected.

It has previously been shown that IGF1 has anabolic effects (7), and combined with GH reduces proteolysis (17). No major improvements in body composition, except for smaller changes in SF and percent FFM in the group treated with GH and IGF1, were recorded in our obese patients. It is well-known that GH treatment leads to water retention (1, 4), which could be part of the increase in FFM. TBW was included in the bioimpedance measurements and no changes were seen within or between groups during the 6-month study. Thus, an increase in TBW does not seem to be the explanation for the change in FFM.

However, all but three patients were previously treated with GH, and therefore the positive impact on body composition might already have occurred. On the other hand, we were interested in the additive effect of IGF1 and major significant differences would have been revealed. Although the study participants included women and men with different types of pituitary diseases, overweight and obese, and different degrees of GHD, any dramatic differences would have been expected to be discovered with the randomised design. However, the strength of our study is the double-blind placebo-controlled design. In a wider context it can be speculated whether the combined treatment with GH and IGF1 or GH alone can be related to the future risk of developing type 2 diabetes, as our results might indicate. To answer this question, further studies with a different study design are needed.

In conclusion, in these patients with GHD and type 2 diabetes, positive effects on insulin sensitivity, SF and percent FFM were recorded in the patients receiving the combined treatment with GH and IGF1 for 6 months. The treatment was well tolerated and no safety issues occurred. Effects of the combined treatment were positive, but limited, and furthermore, taking into account the need of an extra daily injection, it might be an advantageous treatment option in a few selected patients only.

Declaration of interest

C Barner has nothing to disclose. M Petersson is member of the Swedish KIMS study advisory board. B Edén Engström is member of the Swedish KIMS study advisory board and has received lecture fees from Pfizer, Novartis and IPSEN. C Höybye is member of the Swedish KIMS study advisory board and Novo Nordisk study advisory board, and has received lecture fees from Pfizer, Novo Nordisk, and Otsuka and research grants from Pfizer, Novo Nordisk and IPSEN Sweden.

Funding

This trial was supported by grants from Karolinska Institutet, Stockholm, Sweden and IPSEN Sweden.

Acknowledgements

This trial was supported by grants from Karolinska Institutet, Stockholm, Sweden and IPSEN Sweden.

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Received 5 June 2012

Revised version received 27 August 2012

Accepted 3 September 2012