CLINICAL STUDY

The prevalence of the metabolic syndrome is increased in patients with GH deficiency, irrespective of long-term substitution with recombinant human GH

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Abstract

Objectives: Many reports demonstrate improvements in cardiovascular risk factors during GH replacement (rhGH) in adult GH deficiency (GHD). However, it remains to be determined to what extent these changes translate into a reduction of increased cardiovascular morbidity and mortality. The aim of this study was to evaluate the effects of long-term rhGH replacement on the prevalence of the metabolic syndrome (MS).

Design, settings, main outcome measures: The MS was scored by the National Cholesterol Education Program-Adult Treatment Panel III definition in 50 consecutive GHD patients (45 ± 9 years of age), before and after 2 and 5 years of rhGH replacement, and the data of untreated patients were compared with the general population using data from a Dutch population-based study ($n = 1062$, 44 ± 8 years of age).

Results: Hypertriglyceridaemia (46.0 vs 18.5%, $P < 0.0001$), hypertension (66.0 vs 35.5%, $P < 0.0001$) and abdominal obesity (38.0 vs 23.4%, $P = 0.0178$) were more prevalent in untreated patients when compared with controls, resulting in a higher prevalence of the MS in patients (38.0 vs 15.7%, $P < 0.0001$). During rhGH replacement at a mean dose of 0.5 ± 0.2 mg/day resulting in IGF-I concentrations in the normal age-adjusted reference range, mean high-density lipoprotein cholesterol level increased compared with baseline ($P < 0.001$). However, the prevalence of (components of) the MS did not change after 2 or 5 years of treatment with rhGH.

Conclusion: In this study, the prevalence of the MS in patients with GHD is increased compared with healthy controls, irrespective of rhGH replacement.

European Journal of Endocrinology 156 455–462

Introduction

Many reports have documented the beneficial effects of short- (1) and long-term (Table 1) recombinant human growth hormone (rhGH) on cardiovascular risk factors in adults with GH deficiency (GHD). In placebo-controlled studies, with a duration ranging from 1 week to 18 months, rhGH treatment was beneficial for lean and fat body mass, total and high-density lipoprotein (HDL) cholesterol levels and diastolic blood pressure (DBP), whereas rhGH negatively influenced plasma glucose and insulin levels (1). Long-term studies have revealed an increase in HDL cholesterol (2–4) and fasting glucose levels (3–6), and a decrease in triglyceride (TG) levels (3, 7). Systolic blood pressure (SBP) remained unchanged (3, 7, 8), whereas DBP decreased only in one study (8). The actual data reported in these studies indicate that despite the beneficial effects, cardiovascular risk factors remain increased in many patients.

The metabolic syndrome (MS) is a cluster of metabolic abnormalities that identifies persons at high risk for cardiovascular disease (9–11). The aim of this study was to characterize the baseline characteristics of the MS in a cohort of adults with GHD and to evaluate the effect of subsequent rhGH replacement during 5 years on the prevalence of the MS. We hypothesized that long-term rhGH replacement in adults with GHD would ultimately lead to an improved cardiovascular risk profile as assessed by the criteria of the MS.

Methods

Patients

From October 1994 to April 2000, 64 consecutive patients with adult-onset GHD aged 30–59 years were enrolled. In 50 patients, we could score the presence of the MS at baseline and after 5 years of rhGH

Table 1 Observational studies on the effect of recombinant human growth hormone (rhGH) on cardiovascular parameters with a duration of at least 2 years.

Authors	Duration (years)	Numbers (men)	Age	Daily dose	HDL	TG	Glucose	SBP	DBP	Waist circ.	BMI	WH ratio
Colao (7)	2	20 Women	18–45	0.077 mg/kg		↓		=	=		=	
Colao (7)	2	18 Men	19–45	0.065 mg/kg		↓		=	=		↓	
Florakis (5)	2	24 (10)	48	0.4 mg		=	↑					
O'Neal (6)	2	22 (16)	42	0.01 mg/kg	=	=	↑				↑	↓
Garry (26)	3	21 (16)	45.9	0.007 mg/kg	=	=	=					
Al-Shoumer (27)	4	13 (7)	Median 47	0.008 mg/kg	=	=	=					
Götherström (3)	5	118 (70)	49	0.48 mg	↑	↓	↑	=	=		↑	
Present study	5	50 (24)	45	0.5 mg	↑	=	=	=	=	=	↑	=
Svensson (4)	7	11 (7)	48	0.61 mg	↑	=	=	=	=	=	=	
Chrisoulidou (8)	7.1	12 (6)	52	0.7 mg	=	=	=	=	↓	=	↑	
Gibney (2)	10	10 (7)	38	0.008 mg/kg	↑	=						

Observational studies on the effect of rhGH on cardiovascular parameters with a duration of at least 2 years. HDL, high-density lipoprotein cholesterol; TG, triglycerides; SBP, systolic blood pressure; DBP, diastolic blood pressure; Waist circ., waist circumference; TC, total cholesterol; BMI, body mass index; WH ratio, waist-to-hip ratio; ↓, significant decrease; ↑, significant increase; =, no change.

replacement, according to the National Cholesterol Education Program-Adult Treatment Panel III (NCEP-ATP III) definition (12). The other 14 patients were excluded from analysis because insufficient data were available to score the prevalence of the MS ($n=7$) or because rhGH treatment was ended earlier by the patients due to subjective lack of benefit ($n=4$) or side effects ($n=3$). No differences were present between the 14 excluded patients and the other 50 patients with respect to age, gender and body mass index (BMI).

Treatment protocol

Patients were prospectively enrolled in an open-label treatment protocol. GHD was confirmed in all patients by an insulin tolerance test (nadir blood glucose <2.2 mmol/l) with a peak GH concentration <3 μ g/l. After initial measurements were obtained, all patients were treated with s.c. injections of rhGH (Genotropin Pharmacia/Pfizer, Zomacton Ferring, Norditropin Novo Nordisk, or Humatrope Lilly), every evening. The initial dose of rhGH was 0.2 mg/day, which was individually adjusted each month in the first half year to achieve physiological serum insulin-like growth factor-I (IGF-I) concentrations within the age-dependent laboratory reference range IGF-I SDS. Thereafter, this individualized dose was continued in each patient and adjusted, if necessary, to maintain a normal IGF-I concentration for the duration of the study.

Patients with a functioning adenoma (eight patients with Cushing's disease, two patients with acromegaly and seven patients with prolactinoma) were in long-term remission before entering the study. When secondary amenorrhoea was present for more than one year, premenopausal women were defined as luteinizing hormone/follicle-stimulating hormone (LH/FSH) deficient. In men, LH/FSH deficiency was defined as a testosterone level below the reference range (8.0 nmol/l). Thyroid-stimulating hormone (TSH) deficiency was defined as a total or free T_4 level below the reference range. Adrenocorticotrophin (ACTH) deficiency was

defined as an insufficient increase in cortisol levels (absolute value <0.55 μ mol/l) after a corticotrophin-releasing hormone stimulation test or insulin tolerance test. Patients were then treated with hydrocortisone ($n=39$, mean dosage 25 ± 8 mg/day, range 10–40 mg/day), L-thyroxine ($n=44$), testosterone ($n=21$) and/or oestrogen in combination with progestins in premenopausal women only ($n=18$). Conventional substitution therapy was monitored during substitution with rhGH and the respective dosages were adjusted, as required for normalization of clinical and biochemical parameters of pituitary deficiencies. Regular screening for pituitary deficiencies was continued during follow-up. After baseline, additional L-thyroxine substitution was started in one patient and additional hydrocortisone was started in one patient. Patients were treated with antihypertensive and lipid-lowering medication when required following standard patient care procedures. During follow-up, lipid-lowering treatment was initiated by the treating physician in 4 out of the 50 patients, and antihypertensive medication in 6 out of the 50 patients because of additional cardiovascular risk factors.

The study protocol was approved by the local ethics committees. All adult patients gave written informed consent to participation in the study.

Study parameters

The following study parameters were assessed before and after 2 and 5 years of substitution with rhGH: body weight and height, waist circumference, hip circumference, SBP and DBP respectively, and fasting serum levels of glucose, HDL and TG were measured. BMI and waist-to-hip (WH) ratio were calculated. Body weight was measured to the nearest 0.1 kg, and body height was measured barefoot to the nearest 0.001 m. The BMI was calculated as weight in kilograms divided by the square of height in meters. SBP and DBP were measured using the sphygmomanometric cuff method.

Patients were requested to fast overnight before blood samples were taken for laboratory measurements of lipid profiles, glucose and IGF-I concentrations.

The serum IGF-I (nmol/l) concentration was measured by RIA (INCSTAR Corp., Stillwater, MN, USA) after extraction and purification on octadecasilyl-silica columns. The detection limit of this assay is 1.5 nmol/l, and the intra- and inter-assay coefficients of variation were below 11%. Age- and gender-adjusted IGF-I data were determined in the same laboratory (13, 14). IGF-I was expressed as SDS for age- and gender-related normal levels.

GH concentrations in the samples of the insulin tolerance test were measured by time resolved immunofluorometric assay (Wallac, Inc., Turku, Finland). Human biosynthetic GH (Pharmacia and Upjohn, Inc., Uppsala, Sweden) was used as standard, calibrated against WHO-IRP 80-505. The detection limit of this GH assay is 0.01 µg/l with an inter-assay coefficient of variation of 1.6–8.4%, between 0.1 and 15 µg/l.

A Hitachi 747 autoanalyzer (Roche) was used to quantify serum concentrations of glucose and TG. HDL was measured with a homogenous enzymatic assay (Hitachi 911, Roche). In 2003, the Hitachi 747 was replaced by a modular P 800 with no change in the chemistry components.

Control subjects

Control subjects were participants from the monitoring project on risk factors for chronic disease (MORGEN study) organized by the National Institute for Public Health and the Environment (RIVM) (15, 16). From 1993 to 1997, age- and sex-stratified random samples were drawn from the municipality registries from three towns in the Netherlands in a cross-sectional study design. The patients were of the same geographical area as the controls. Anthropometry, BP and total and HDL cholesterol levels were measured during the survey. Stored fasting plasma samples from subjects participating in the period 1993–1995 were retrieved in 1996 and analysed for TG concentrations ($n=1378$). All participants aged 30 years and older were included in the present study ($n=1062$). Since some of our patients were recruited somewhat later than the controls were studied, we investigated whether time frame of recruitment in our patients influenced the prevalence of the MS. We compared the prevalence of the MS in patients who started rhGH replacement exactly during the time in which controls were selected ($n=31$), with patients who started after the time in which controls were selected ($n=19$) and found no significant difference in the prevalence of the MS (32 vs 43%, $P=0.285$).

Standard enzymatic methods were used to measure HDL cholesterol and glucose levels. TG concentrations were measured with Abbott Spectrum clinical analyzer

(Abbott Laboratories). BP was measured by random sphygmomanometer.

Definition of the MS

The MS was scored according to the definition of the NCEP-ATP III published in 2001 (12), which defines the MS as the presence of three or more of the following criteria:

- 1) fasting plasma glucose concentration ≥ 6.1 mmol/l;
- 2) TG concentration ≥ 1.69 mmol/l;
- 3) HDL concentration <1.04 mmol/l in men and <1.29 mmol/l in women;
- 4) BP $\geq 130/85$ mmHg;
- 5) waist circumference >102 cm in men and >88 cm in women.

Statistical analysis

Statistical analysis was performed using SAS for Windows, version 9.1 and SPSS for Windows, version 12.0 (SPSS, Inc., Chicago, IL, USA). Results are expressed as mean $\pm$ s.d. unless specified otherwise. Paired samples *t*-tests or ANOVA with repeated measurements with Bonferroni correction for multiple comparisons were used to assess the differences of continuous variables, when appropriate. χ^2 tests were used to assess the difference in prevalence of the MS between patients and controls. Friedman test for related fractions was used to assess the effect of treatment on the prevalence of the MS. Logistic regression was used to identify indicators of the prevalence of the MS at baseline. General linear model for repeated measurements was used to assess the prevalence of the MS at baseline and after 5 years of treatment taking trend in BMI into account. $P<0.05$ was considered to be significant.

Results

Untreated GHD adults compared with controls

Characteristics of patients and controls Gender was equally distributed in both cohorts. There were no differences in age, gender and BMI between controls and patients. Patients mean maximal GH response during insulin-induced hypoglycaemia was only 0.3 ± 0.4 µg/l (range 0.1–1.6 µg/l), confirming the diagnosis of severe GHD (17) (Table 2).

Cardiovascular risk factors Patients had significantly lower fasting glucose concentrations and significantly higher mean TG, SBP, DBP, waist circumference in men, and WH ratio in men and women compared with controls (Table 3).

Table 2 Characteristics of patients with adult-onset GH deficiency (GHD) and controls.

		Patients (n=50)	Controls (n=1062)	P value
Gender (%)	Male/female	48/52	52/48	NS
Age (years (mean $\pm$ s.d.))		45.2 $\pm$ 9.1	43.8 $\pm$ 8.0	NS
BMI (kg/m ² (mean $\pm$ s.d.))		26.7 $\pm$ 4.2	25.7 $\pm$ 4.0	NS
Antihypertensive medication (%)		12	17	NS
Lipid-lowering drugs (%)		10	13	NS
Etiology of GHD (%)	Non-functioning pituitary adenoma	24		
	Functioning pituitary adenoma*	34		
	Craniopharyngioma	14		
	Other [†]	28		
Surgery (%)		78		
Radiotherapy (%)		56		
ADH deficiency (%)		30		
ACTH deficiency (%)		78		
TSH deficiency (%)		88		
LH/FSH deficiency (%)	Men	88		
	Women	100		

Data are presented as percentages unless specified otherwise. *Prolactinoma, adrenocorticotrophin (ACTH)-producing adenoma, GH-producing adenoma. [†]Trauma, hypophysitis, germinoma, epidermoid cyst, Sheehan's syndrome, unknown cause. NS, not significant.

Comparison of components of the MS Hypertriglyceridaemia (46.0 vs 18.5%, $P < 0.0001$), hypertension (66.0 vs 35.5%, $P < 0.0001$) and abdominal obesity (38.0 vs 23.4%, $P = 0.0178$) were significantly more prevalent in patients compared with controls (Fig. 1).

Comparison of the MS The prevalence of the MS was 38% (19/50 patients) vs 15.7% in controls ($P < 0.0001$). In patients, BMI was a significant indicator of the presence of the MS (odds ratio 1.2, 95% CI (1.0–1.4), $P = 0.031$). The presence of the MS was not dependent on age, gender, aetiological diagnosis of GHD, surgery or radiotherapy ($P = 0.823$, 0.729, 0.340, 0.704, 0.093 respectively). There were no correlations between the calendar time of diagnosis of GHD and the

prevalence of the MS or between the interval between diagnosis of GHD and start of rhGH replacement (Fig. 1).

Effects of long-term substitution with rhGH

rhGH substitution characteristics After one year of rhGH treatment, a stable dose of rhGH was maintained and the mean dose after 5 years was 0.5 ± 0.2 mg/day. Mean IGF-I SDS were -2.0 ± 0.8 at baseline, -0.2 ± 1.8 after 2 years and 0.8 ± 2.0 after 5 years of rhGH replacement ($P < 0.001$; Table 3).

Effects of rhGH on cardiovascular risk factors

Fasting glucose and TG levels remained unchanged during follow-up, whereas mean HDL levels increased

Table 3 Metabolic and anthropometric parameters in adult-onset GH-deficient patients after 5-year treatment.

		Baseline controls (n=1062)	Baseline patients (n=50)	Two-year treatment patients	Five-year treatment patients	P values between patients (baseline) and controls	P values between patients baseline and 2-year treatment	P values between baseline and 5-year treatment
IGF-I SDS			-2.0 ± 0.8	-0.2 ± 1.8	0.8 ± 1.9		< 0.001	0.001
Glucose (mmol/l)		5.4 ± 1.1	4.8 ± 1.2	5.0 ± 0.7	5.3 ± 1.3	< 0.0001	NS	NS
HDL (mmol/l)	Men	1.2 ± 0.3	1.1 ± 0.3	1.2 ± 0.3	1.3 ± 0.4	NS	NS	0.081
	Women	1.5 ± 0.4	1.5 ± 0.6	1.6 ± 0.5	1.7 ± 0.6	NS	NS	0.020
TG (mmol/l)		1.3 ± 1.1	1.9 ± 1.1	2.0 ± 1.0	2.1 ± 1.4	< 0.0001	NS	NS
Systolic BP (mmHg)		121 ± 16	128 ± 12	137 ± 19	129 ± 14	0.0004	0.017	NS
Diastolic BP (mmHg)		79 ± 10	83 ± 8	86 ± 8	82 ± 9	0.0045	NS	NS
Waist circumference (cm)	Men	94.2 ± 10.8	101.5 ± 8.8	98.9 ± 8.3	104.3 ± 9.8	0.0017	NS	0.062
	Women	82.7 ± 11.3	86.8 ± 12.0	87.8 ± 13.3	88.4 ± 12.3	NS	NS	NS
WH ratio	Men	0.91 ± 0.07	0.98 ± 0.05	0.95 ± 0.05	0.99 ± 0.06	< 0.0001	0.008	0.042
	Women	0.80 ± 0.07	0.86 ± 0.07	0.85 ± 0.06	0.87 ± 0.06	< 0.0001	NS	NS
BMI (kg/m ²)		25.7 ± 4.0	26.7 ± 4.2	26.7 ± 4.1	27.8 ± 4.7	NS	NS	0.002

Values are expressed as mean $\pm$ s.d. Insulin-like growth factor-I (IGF-I) SDS, IGF-I standard deviation scores; HDL, high-density lipoprotein cholesterol; TG, triglycerides; BP, blood pressure; WH ratio, waist-to-hip ratio; BMI, body mass index.

Figure 1 The prevalence of (components of) the metabolic syndrome in patients with growth hormone deficiency at baseline compared with healthy controls. Black bars denote GHD patients at baseline and white bars denote controls. The prevalence of hypertriglyceridaemia, hypertension and abdominal obesity was significantly higher in GHD adults compared with controls (* $P < 0.0001$, † $P < 0.02$). The prevalence of the metabolic syndrome was significantly higher in the total cohort (38.0 vs 15.7%, * $P < 0.0001$). TG, triglycerides; HDL, high-density lipoprotein cholesterol; BP, blood pressure; Waist, waist circumference; MS, metabolic syndrome.

during follow-up from 1.3 ± 0.5 mmol/l at baseline to 1.4 ± 0.5 mmol/l after 2 years and to 1.5 ± 0.5 mmol/l after 5 years of rhGH treatment ($P < 0.001$). However, when men and women were analysed separately the increase in HDL in men failed to reach statistical significance ($P = 0.081$). SBP and DBP remained unchanged, except for a transient increase in SBP during the first 2 years of treatment (128 ± 12 mmHg at baseline, 137 ± 19 mmHg after 2 years ($P = 0.017$) and 129 ± 14 mmHg after 5 years of treatment). Mean BMI increased from 26.7 ± 4.2 to 27.8 ± 4.7 kg/m² ($P = 0.002$). Mean waist circumference remained unchanged, except for a slight increase in men after 5 years of treatment which failed to reach statistical significance ($P = 0.062$). This increase was also reflected in the increase in WH ratio seen only in men during follow-up (0.98 ± 0.05 at baseline to 0.99 ± 0.06 after 5 years of treatment, $P = 0.042$; Table 3).

Effects of rhGH on components of the MS After 2 and 5 years of rhGH replacement, the prevalence of the components of the MS remained unchanged. The decrease in prevalence of low HDL levels (38% at baseline, 42% after 2 years to 20% after 5 years) failed to reach statistical significance ($P = 0.068$; Fig. 2, Table 4).

Figure 2 The prevalence of (components of) the metabolic syndrome in patients with growth hormone deficiency at baseline and after 2 and 5 years of substitution of rhGH. Black bars denote GHD patients at baseline; white bars denote patients after 2 years of substitution with rhGH and grey bars denote patients after 5 years of substitution with rhGH. The prevalence of an abnormal low HDL decreases after 5 years of substitution with rhGH (* $P = 0.068$).

Effects of rhGH on the prevalence of the MS After 2 years of rhGH treatment, the prevalence of the MS was 42% and after 5 years the MS was still present in 19 patients (38.0%, not significant in a Friedman test for related fractions; Fig. 2).

Influence of age, gender, BMI and aetiological diagnosis of GHD When adjusted for the increase in BMI during the period, the prevalence estimate for the MS did not change during 5 years of treatment with rhGH (39.6% at baseline vs 36.4% after 5 years, $P = 0.71$).

There were no differences in age, gender, BMI, aetiological diagnosis of GHD, and applied treatments (radiotherapy or surgery), mean dose of rhGH after 5

Table 4 Prevalence of components of the metabolic syndrome in adult-onset GH-deficient adults before and after 2 and 5 years of treatment.

	Baseline	Two-year treatment	Five-year treatment	P values treatment*
Increased fasting glucose (%)	8.0	6.1	10.0	0.717
Increased BP (%)	66.0	64.6	65.3	0.913
Decreased HDL cholesterol (%)	38.0	35.6	20.0	0.068
Increased TG levels (%)	46.0	57.8	58.0	0.084
Increased waist circumference (%)	38.0	43.2	57.4	0.741
Metabolic syndrome (%)	38.0	41.9	38.0	0.846

Values are expressed as percentages. BP, blood pressure; HDL, high-density lipoprotein cholesterol; TG, triglycerides; *Friedman test for related fractions.

years and mean change in IGF-I levels between patients who acquired the MS ($n=9$), who were relieved from the MS ($n=9$) or who were stable during follow-up (MS present ($n=10$) or absent ($n=22$)).

One of the eight patients with Cushing's disease had the MS at baseline and after 5 years none of the patients treated for Cushing's disease had the MS.

There were no significant differences in the prevalence of the MS at any time point between patients with or without ACTH deficiency and patients with or without thyroid stimulating hormone deficiency, or between patients with or without LH/FSH deficiency. Logistic regression revealed no statistically significant influence of increasing number of anterior pituitary deficiency or increasing hydrocortisone dose on the prevalence of the MS at any time point during follow-up.

Discussion

In this study, patients with GHD have a more than twofold increased prevalence of the MS when compared with the general population. This increase is predominantly due to increased prevalence of hypertension, abdominal obesity and hypertriglyceridaemia among patients with GHD. Long-term rhGH treatment increases HDL cholesterol concentrations, whereas fasting glucose and TG concentrations remained unchanged in accordance with previous observations (references summarized in Table 1). However, rhGH treatment did not reduce the high prevalence of other cardiovascular risk factors assessed by the criteria of the MS.

The rationale for treatment with rhGH is to improve the metabolic abnormalities and psychological well-being, with the expectation to, ultimately, normalize life expectancy. The results of placebo-controlled trials with rhGH on cardiovascular risk factors were recently evaluated in a meta-analysis (1). In those studies (with a duration ranging from 1 week to 18 months), rhGH treatment was beneficial for lean and fat body mass, total and HDL cholesterol levels and DBP, but negatively influenced plasma glucose and insulin levels (1). Moreover, the weighted mean change, especially in total and low density lipoprotein (LDL) cholesterol levels and DBP, was restricted to a maximum of only 0.3 mmol/l, 0.5 mmol/l and 1.8 mmHg respectively (1). It remains unclear to what extent these relatively small changes would translate into a net beneficial effect on cardiovascular risk.

Some factors may have influenced the effect of rhGH in our study. The data obtained after five years of follow-up were not only affected by rhGH treatment, but also by age and BMI. In our control population, an increase of five years in age was associated with a significant increase in the prevalence of the MS from 15.7 to 20.9% (data not shown). However, this does not affect our conclusions with respect to the limited effects of rhGH treatment, since the prevalence of the MS remained

much higher in GHD patients, irrespective of the duration of follow-up and rhGH treatment. Additional adjustment for the trend in BMI did not affect our conclusions. Moreover, after 2 years of rhGH replacement, in which the BMI did not increase, the prevalence also remained unaffected by rhGH replacement. However, we cannot exclude the possibility that rhGH replacement was able to stabilize the prevalence of the MS and prevented an increase in prevalence of cardiovascular risk factors, associated with the observed increase in BMI and age. Although BMI significantly increased during follow-up, waist circumference and WH ratio remained unchanged. The increase in BMI in patients receiving rhGH replacement for five years is consistent with a previous reports in GHD adults (3, 6, 8). Moreover, with linear regression in our control population, an increase in BMI of 0.5 kg/m² after a 5-year age increase was estimated. Thus, it appears that increasing age of GHD adults is associated with an increase in BMI, irrespective of rhGH replacement, just as in the normal population.

Waist circumference transiently decreased in men during follow-up, but remained unchanged after five years of rhGH replacement when compared with baseline. This is in line with findings of large short-term studies by Filipsson *et al.* and of Abs *et al.* in which waist circumference decreased after one and two years of rhGH treatment respectively (18, 19), as well as with findings of one long-term study in which waist circumference was unchanged after seven years of rhGH replacement (8). Thus, it seems that short-term rhGH replacement is able to induce favourable changes in waist circumference but unable to prevent the increase in waist circumference due to ageing as is seen in healthy adults.

The high prevalence of the MS could also be related to the complex syndrome of anterior pituitary deficiencies. All patients received adequate replacement therapies for pituitary insufficiencies prior to the start of study except for GHD. Moreover, conventional substitution therapy was carefully monitored during substitution with rhGH and the respective dosages were adjusted as required for normalization of clinical and biochemical parameters of pituitary deficiencies. The prevalence of the anterior pituitary deficiencies in our cohort is comparable with the prevalence found in a large study of long-term rhGH replacement in GHD adults by Götherström *et al.* (3). In our study, 94% of patients had a deficiency of at least one anterior pituitary hormone compared with 92% in the study of Götherström *et al.* and 78% in our study had at least three other deficiencies besides GHD compared with 68% in the study of Götherström *et al.* (3). Although the prevalence of pituitary deficiencies might influence the prevalence of the MS in these patients, we did not find any differences between patients with or without various deficiencies. This might be related to the limited number of patients included in the present study.

Recently, it has been shown by Filipsson *et al.* that dose of glucocorticoids in the treatment of ACTH deficiency influences BMI, TG, LDL and total cholesterol levels in a dose-dependent manner, whereas it did not affect treatment response to rhGH replacement (18). In our limited number of patients and limited range of hydrocortisone doses used, we were unable to show such a relationship between the MS and hydrocortisone dose.

The underlying diseases that caused GHD could also influence our results. Patients with Cushing's disease are known to have a high prevalence of cardiovascular risk factors, even after successful long-term cure of Cushing's disease (20). On the other hand, Feldt-Rasmussen *et al.* found no differences in WH ratio, BMI, TG, and total, LDL and HDL cholesterol levels between patients with GHD due to previous treatment for Cushing's disease compared with patients with GHD due to other aetiologies (21). In our limited number of patients, we were unable to demonstrate differences in the prevalence of the MS between patients treated for Cushing's disease or acromegaly and patients treated for non-functioning pituitary adenomas, or between patients with producing pituitary adenomas compared with patients with non-functioning adenomas.

In our study, in four patients, lipid-lowering treatment was started during follow-up and in six patients, antihypertensive medication. The exclusion of these patients from the analysis of lipid parameters or systolic and DBP measurements respectively did not affect our observations. Recently, Grundy *et al.* (22) proposed to take the medication used into account. We therefore chose to perform a second analysis in which we also scored antihypertensive and lipid-lowering medication in patients and controls. The comparison with controls remained unchanged (38 vs 16.5%, $P < 0.0001$) as well as the prevalence after 5 years of substitution with rhGH (38% at baseline vs 43% after 5 years, NS vs baseline).

The concept of the MS is subject to debate, because the pathophysiological basis of the proposed syndrome is unclear (23), and because the combination of cardiovascular risk factors does not add to the risk related to the individual risk factors (24). Nonetheless, this current debate does not affect our conclusions because we also focus on the prevalence of the individual, well-recognized cardiovascular risk factors, which have all individually been associated with increased cardiovascular morbidity and mortality in the general population (9, 10, 24).

It needs to be established whether adult GHD patients with the MS have the same risks for cardiovascular morbidity and mortality compared with those with the MS in the general population. It remains to be studied whether the prognostic significance of the MS in these patients with GHD is the same as in the healthy general population. Moreover, the pathogenesis of the MS might be different in our patients compared with the general population. For example, the effect of GHD and rhGH replacement on insulin sensitivity might influence the

prevalence of the MS in our patients. Data so far on insulin resistance during rhGH replacement are conflicting, but some studies have pointed towards an improved insulin sensitivity during long-term rhGH replacement (4), which could be attributed to favourable changes in body composition (25). Furthermore, it remains to be studied in prospective trials if GHD adults may benefit from more aggressive antihypertensive and lipid-lowering therapy and lifestyle intervention to reverse the metabolic abnormalities seen in the adult GHD syndrome.

In conclusion, the prevalence of the MS in our GHD adults is significantly higher compared with the general population, irrespective of rhGH treatment. Apparently, appropriate substitution of rhGH and other hormones in adult patients with GHD is insufficient to improve this adverse cardiovascular risk profile.

Acknowledgements

The MORGEN project was financially supported by the Ministry of Public Health, Welfare and Sports of The Netherlands and the National Institute for Public Health and the Environment. The sponsors did not have a role in the study design, collection/analysis/interpretation of the data, writing of the report or the decision to submit the paper for publication.

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Received 23 November 2006

Accepted 5 February 2007