A Reappraisal of Diagnosing GH Deficiency in Adults: Role of Gender, Age, Waist Circumference, and Body Mass Index

Annamaria Colao, Carolina Di Somma, Silvia Savastano, Francesca Rota, Maria Cristina Savanelli, Gianluca Aimaretti, and Gaetano Lombardi

Department of Molecular and Clinical Endocrinology and Oncology (A.C., C.D.S., S.S., F.R., M.C.S., G.L.), Section of Endocrinology, University of Naples "Federico II," Italy, 80131 Naples, Italy; and Section of Endocrinology (G.A.), Department of Clinical and Experimental Medicine, University "A. Avogadro" del Piemonte Orientale, 28100 Novara, Italy

Objective: The objective of the study was to reevaluate the diagnostic accuracy of GH peak after GHRH plus arginine test (GHRH+ARG) according to patients' age, body mass index (BMI), and waist circumference to diagnose GH deficiency (GHD).

Outcome Measures: GH peak after GHRH+ARG and IGF-I levels reported as SD score.

Subjects: Subjects included 408 controls (218 women, 190 men, aged 15–80 yr) and 374 patients with hypopituitarism (167 women, 207 men, aged 16–83 yr).

Results: In the (elderly) healthy subjects 15–25 yr old (young), 26–65 yr old (adults) and older than 65 yr, GH cutoffs were 15.6, 11.7, and 8.5 μg/liter, 11.8, 8.1, and 5.5 μg/liter, and 9.2, 6.1, and 4.0 μg/liter, respectively, in the lean, overweight, and obese subjects. Waist circumference was the best predictor of GH peak ($t = -7.6$, $P < 0.0001$) followed by BMI ($t = -6.7$, $P < 0.0001$) and age ($t = -5.7$, $P < 0.0001$). Based on the old (<9.1 μg/liter) and new GH cutoff, 286 (76.5%) and 276 (73.8%) of 374 hypopituitary patients had severe GHD. The receiving-operator characteristic analysis showed GH cutoffs in line with the third percentile or slightly higher results so that the prevalence of GHD increased to 90.1%.

Conclusions: The results of the current study show that waist circumference and BMI are the strongest predictors of GH peak after GHRH+ARG followed by age. However, the old cutoff value of 9.0 μg/liter was in line with the new cutoffs in 95% of patients. **(*J Clin Endocrinol Metab* 94: 4414–4422, 2009)**

GH Deficiency (GHD) in the adult population causes a syndrome characterized by altered body composition, reduced bone mineralization, unfavorable lipid profile, reduced cardiac performance, early atherosclerosis, and impaired quality of life (1, 2). Even if not completely proven, it is suggested that these abnormalities are associated with reduced life expectancy (3–8).

Currently several pharmacological stimuli of GH secretion, such as insulin-induced hypoglycemia (ITT), glucagon, GHRH plus arginine (GHRH+ARG), and GHRH plus GH releasing peptide-6, are used to diagnose GHD (9, 10). With currently available GH assay methods, severe GHD in adults is reportedly diagnosed by a GH peak less than 3.0 μg/liter to ITT or glucagon and less than 9.1 μg/liter to GHRH+ARG. In childhood, diagnosis of GHD is based on a peak GH response of less than 5–7 μg/liter to ITT, a cutoff that was validated against height velocity (11). Similarly, the validity of cutoff GH after GHRH+ARG proposed by Aimaretti *et al.* (12) has been supported by a correlation with impairment of lipid pro-

ISSN Print 0021-972X ISSN Online 1945-7197
Printed in U.S.A.

doi: 10.1210/jc.2009-1134 Received May 28, 2009. Accepted August 6, 2009.
First Published Online September 22, 2009

Abbreviations: BMI, Body mass index; GHD, GH deficiency; GHRH+ARG, GHRH plus arginine; HDL, high-density lipoprotein; ITT, insulin-induced hypoglycemia; ROC, receiving-operator characteristic; SDS, SD score.

file (13), bone loss (14), and cardiac abnormalities (15). More recently evidence is accumulating that requires a revision of diagnostic GH cutoffs proposed by the GRS Consensus Statement in 1998 (16) to take into consideration both age and body mass index (BMI) of the patients (17–21). In transition from adolescence to adulthood, between ages 16 and 25 yr, higher cutoff levels are suggested to be necessary to diagnose GHD: GH peak after ITT or GHRH+ARG, respectively, was proposed to be less than 5.0 μg/liter by Clayton *et al.* (17) or less than 6.1 μg/liter by Maghnie *et al.* (18) and less than 19.0 μg/liter by Corneli *et al.* (21). These cutoff values are approximately twice the adult threshold. Conversely, in obese hypopituitary patients, both Biller *et al.* (19) and Corneli *et al.* (20) reported that diagnostic GH cutoff by the GHRH+ARG test should be lowered to 4.1–4.2 μg/liter, thus approximately half the adult threshold.

The validation of GH cutoff after all stimulation tests is essential because in both the United States and Europe, GH replacement is permitted only in patients with severe GHD.

To provide a reappraisal of appropriateness of GHRH+ARG test in diagnosing GHD in patients with organic hypopituitarism, we designed this retrospective study. The first aim of the study was to analyze the peak GH after GHRH+ARG in a large series of healthy controls grouped on the basis of their gender, age, and BMI. Then limits of the first percentile of controls were used to investigate the prevalence of severe GHD in 374 patients with organic hypopituitarism. Because in adults a clinical end point as valid as growth velocity in children is lacking, the GH peak values were analyzed in comparison with IGF-I levels [evaluated as SD score (SDS) from the mean value of controls] and total to high-density lipoprotein (HDL) cholesterol ratio because of the well-known cardiovascular risk of GHD patients (22). The appropriateness of the cutoffs provided by the percentiles analysis were also analyzed by receiving-operator characteristic (ROC) curves to verify the similarity or discordance between these two methods.

Subjects and Methods

Subjects

Between January 1, 1997, and December 31, 2007, the GHRH+ARG test was performed at the Department of Molecular and Clinical Endocrinology and Oncology of the University Federico II (Naples, Italy) in 782 subjects, 374 patients with hypopituitarism, and 408 controls.

Controls

A total of 408 subjects (218 women, 190 men, aged 15–80 yr), recruited among the medical and paramedical personnel of our department and their relatives, and the patients' relatives agreed to serve as controls.

Patients

A total of 374 patients with overt or suspected hypopituitarism total or partial (167 women, 207 men, aged 16–83 yr) were studied. According to our routine procedure (23–25), before undergoing GH testing, the patients were under stable replacement therapy with L-thyroxine (50–150 μg orally daily), cortisone acetate (25–37.5 mg/day), intranasal desmopressin (5–20 μg/d), testosterone-enanthate (250 mg im every 3 wk or monthly) in men, and transdermal estrogens associated with progesterone in premenopausal females, according to individual patients' endocrine status. Adequacy of hormone replacement therapy was periodically assessed by measuring serum-free thyroid hormones, testosterone, urinary free cortisol (if indicated), blood pressure, and serum Na^+ and K^+ measurements. Forty-five patients had received GH treatment during childhood and were retested in adulthood to confirm GHD, in accordance with modern guidelines (17, 26).

All data presented in this study derive from a study protocol dedicated to investigate the effects of GH replacement on the cardiovascular system in patients with GHD compared with controls. This study was approved by the Ethical Committee of the Federico II University of Naples in 1997 (no. 63/97). All subjects gave their informed consent to the study. The profile of patients and controls at study entry is shown in Table 1.

Study design

This was an analytical, retrospective, controlled study to reevaluate GH cutoff after GHRH+ARG to diagnose severe and partial GHD in adults according to gender, age, and BMI. As for age, we considered the following subgroups: 15–25 yr (young), 26–65 yr (adults), and older than 65 yr (elderly). As for BMI we considered the following subgroups: lean, BMI 18–25 kg/m^2; overweight, 25–30 kg/m^2; and obese, greater than 30 kg/m^2. A secondary analysis was planned in the females and males, separately with waist circumference below or above 88 or 102 cm (27). The GHRH+ARG test was chosen because of previous studies showing reliable cutoff for the GHD diagnosis, as correlated with lipid, bone, and cardiac status (13–15) associated with high reproducibility of this test as well (28, 29). Data were validated against IGF-I levels for age and gender, and the total to HDL cholesterol ratio as marker of cardiovascular risk (30). Because no difference was found between female and male controls, the difference according to gender was removed. Additionally, because no lean subjects, a minority of overweight subjects (8%), and all but 17 obese subjects had a waist circumference above the cutoff of 88 and 102 cm, in females and males, respectively, a separate analysis according to waist circumference was not performed.

Study protocol

Anthropometric measurements were performed with the subjects wearing only underwear without shoes. Standing height was measured to the nearest centimeter using a wall-mounted stadiometer. Body weight was determined to the nearest 50 g using a calibrated balance beam scale. BMI was calculated as weight (kilograms) divided by height squared meters. Measurements of the waist circumference were taken at the midpoint between umbilicus and xiphoid. The GHRH+ARG test was per-

TABLE 1. Profile of patients and controls at study entry

	Controls	Patients	*P*
n	408	374	
Women/men	218/190	167/207	
Age (yr)	43.3 ± 21.5	44.1 ± 16.3	0.62
BMI (kg/m^2)	27.8 ± 7.4	26.8 ± 4.9	0.26
Waist circumference (cm)	84.3 ± 17.9	88.1 ± 10.0	<0.0001
Peak GH after GHRH+ARG (μg/liter)	41.9 ± 22.3	8.7 ± 16.7	<0.0001
Serum IGF-I levels (μg/liter)	221.3 ± 77.0	107.3 ± 70.8	<0.0001
IGF-I SDS	0.26 ± 0.77	−1.24 ± 1.01	<0.0001
Total cholesterol levels (mg/dl)	191.0 ± 29.2	219.9 ± 45.8	<0.0001
HDL-cholesterol levels (mg/dl)	58.6 ± 7.0	44.8 ± 10.3	<0.0001
Total to HDL cholesterol ratio	3.34 ± 0.84	5.26 ± 1.93	<0.0001
Cause of hypopituitarism			
Clinically nonfunctioning pituitary adenomas		192	
PRL-secreting adenomas		85	
ACTH-secreting adenoma		6	
GH-secreting adenoma		4	
Craniopharyngiomas		17	
Idiopathic childhood onset		21	
Traumatic brain injury		9	
Primary empty sella		27	
Meningiomas		4	
Dysgerminomas		2	
Pituitary metastastis		2	
Cyst of the Rathke's pouch		5	

PRL, Prolactin.

formed in accordance with Ghigo *et al.* (28). ARG (arginine hydrochloride; Salf, Bergamo, Italy) was given at the dose of 0.5 g/kg, up to a maximal dose of 30 g slowly infused from time 0 to 30 min, whereas GHRH (1–29) (Geref; Serono, Rome, Italy) was given at the dose of 1 μg/kg as iv bolus at time 0. Blood samples were taken every 15 min from 0 up to 90 min. The highest GH levels measured from time 30 to 90 min during the test was taken for analysis as peak GH. Serum IGF-I levels, total cholesterol, HDL cholesterol, triglycerides, glucose, and insulin levels were assayed within 1 wk from the GH testing.

Assays

During the decade span of study performance, in our laboratory the GH assay changed: sensitivity ranged from 0.2 to 0.05 μg/liter. Serum IGF-I was measured by immunoradiometric assay after ethanol extraction; the normal ranges are reported elsewhere (31). The SDS for age and gender was also calculated according to normal IGF-I levels for age and gender (31).

Statistical analysis

The statistical analysis was performed by StatDirect Statistical Software (version 2,6,2 of the 23/04/07, Cheshire, UK, http://www.statsdirect.com/update.htm). Data are shown as mean ± SD unless otherwise specified. The first and third percentiles of GH peak distribution after GHRH+ARG were determined in the control group according to the age and BMI groups. The comparison between patients and controls was performed by the Student's *t* test for unpaired data. For the purpose of this study, the GH peak below or equal to the first percentile was diagnostic of severe GHD and the GH peak between the first and the third percentile was diagnostic of partial GHD, whereas the GH peak above the third percentile was considered to represent a normal GH secretion. The correlation between GH peak after GHRH+ARG in the controls and patients group and sex, age, BMI, waist circumference, smoking, exercise, and pituitary disease (only in the patients group) was analyzed by calculating the Pearson's coefficient. The stepwise multiple regression was then applied to analyze the best predictor of GH peak after GHRH+ARG in the two populations; in this analysis were entered only the variables with a *P* value less than 1%. Because of absence of correlation between gender and GH peak, comparison among different age and BMI groups was performed by the Kruskal-Wallis test followed by the Dunn's test for all pairs of columns; also in this case, the significance across groups was calculated by applying the Bonferroni correction (*P* less than 1%). Then cutoff thresholds for peak GH after GHRH+ARG were analyzed by ROC curves calculated using MedCalc Software for Windows (MedCalc, Mariakerke, Belgium) in the patients *vs.* controls, according to the different age and BMI categories. Data are expressed as sensitivity and specificity with their 95% confidence intervals in parentheses. The prevalence of GHD according to GH cutoff values derived from percentiles and ROC analysis were compared to verify which method was more reliable to use for diagnosis.

Results

The control group

No difference was found between male and female subjects (41.2 ± 19.8 *vs.* 42.6 ± 24.3 μg/liter, $P = 0.91$). The young (n = 135; 52.4 ± 24.1 μg/liter) had higher GH peak than the adults (n = 166; 40.0 ± 20.2 μg/liter, $P < 0.0001$) and the elderly (n = 107; 31.5 ± 17.0 μg/liter, $P < 0.0001$), and the adults had higher GH peak than the el-

derly ($P = 0.0011$). Similarly, the obese (n = 107; 21.1 ± 10.1 μg/liter) had lower GH peak than the lean (n = 173; 56.4 ± 20.0 μg/liter, $P < 0.0001$) and the overweight (n = 126; 39.8 ± 17.6 μg/liter, $P < 0.0001$), whereas the lean had higher GH peak than the overweight subjects ($P < 0.0001$). Within individual BMI classes, among the lean subjects, higher GH peaks were found in the young *vs.* the adults and the elderly, and in the adults *vs.* the elderly, among the overweight and the obese subjects, higher GH peaks were found in the young *vs.* the adults and the elderly (Table 2). The percentiles of GH peak distribution considered by age and BMI are shown in Table 2. The waist circumference was the best predictor of GH peak after GHRH+ARG (t = −7.6, $P < 0.0001$) followed by BMI (t = −6.7, $P < 0.0001$) and age (t = −5.7, $P < 0.0001$). The GH peak after GHRH+ARG was significantly and positively correlated with IGF-I SDS (r = 0.45, $P < 0.0001$; Fig. 1) and inversely correlated with the total to HDL cholesterol ratio (r = −0.66, $P < 0.0001$; Fig. 1).

The patient group

Table 3 shows the distribution of GH peak in young, adult, elderly, lean, overweight, and obese patients. Similar to controls, the GH peak after GHRH+ARG in the patients was significantly correlated with age (r = −0.12, $P = 0.023$), gender (r = −0.12, $P = 0.02$), waist circumference (r = −0.32, $P < 0.0001$), BMI (r = −0.22, $P < 0.0001$), IGF-I SDS, and total to cholesterol ratio (Fig. 1). The major predictor of GH peak after GHRH+ARG in the patient group was waist circumference (t = −5.48, $P < 0.0001$) followed by BMI (t = −2.30, $P = 0.022$).

Based on the GH peak after GHRH+ARG less than 9.1 μg/liter as diagnostic of severe GHD (28), 286 of 374 hypopituitary patients (76.5%) had severe GHD. As compared with patients with a normal GH secretion, in severe GHD patients, the IGF-I SDS was −1.66 ± 0.67 *vs.* 0.08 ± 0.71 ($P < 0.0001$). Overall, an IGF-I SDS of 2 or less was found in 89 patients (23.8%), all of them had severe GHD and only five of them had isolated GHD (5.6%). By applying the different limits of first percentiles of GH peak according with age and BMI, we found that the diagnostic accuracy did not change significantly compared with the old one (73.8%; Table 3). In severe GHD (n = 282), the IGF-I SDS was −1.65 ± 0.68 *vs.* non-GHD patients (n = 92, −0.01 ± 0.77; $P < 0.0001$). Only 19 patients (5.1%) received a wrong diagnosis with the old cutoff: eight of them did not receive a correct diagnosis of severe GHD, whereas 11 patients were misdiagnosed as severe GHD. The GH peak was lower in patients with two or more deficits than in those with one deficit or isolated GHD (supplemental Table, published as supplemental data on The Endocrine Society's Journals Online web site at http://

TABLE 2. GH peak after GHRH+ARG in the 382 controls grouped according to age and BMI

	Young (15–25 yr)				Adults (26–65 yr)				Elderly (>65 yr)				
	n		1 C	3 C	n		1 C	3 C	n		1 C	3 C	*P*
Lean (L)	62	68.5 ± 18.7 (63.7–73.2)[a,b]	15.6	21.3	73	53.3 ± 17.7 (49.1–57.4)[c]	11.8	21.5	38	42.6 ± 15.0 (37.7–47.5)	9.2	13.5	<0.0001
Overweight (OW)	36	50.8 ± 18.8 (44.4–57.1)[a,b]	11.7	16.5	53	38.0 ± 14.5 (34.0–42.0)	8.1	15.4	37	31.6 ± 15.6 (26.4–36.7)	6.1	9.8	0.0025
Obese (OB)	37	27.1 ± 10.9 (23.5–30.7)[c]	8.5	13.4	40	18.4 ± 7.6 (16.0–20.8)	5.5	9.5	32	14.6 ± 9.3 (11.3–17.9)	4.0	5.0	0.012
	135				166				107				
P^1	0.013 L *vs.* OW 0.33 L *vs.* OB 0.022 OW *vs.* OB 0.0041				0.0002 L *vs.* OW 0.041 L *vs.* OB <0.0001 OW *vs.* OB 0.026				0.033 L *vs.* OW 0.34 L *vs.* OB 0.012 OW *vs.* OB 0.026				

Data are shown as mean ± SD and 95% confidence interval in *parentheses*. 1 C, First percentile; 3 C, Third percentile. *P* refers to the results of the Kruskal-Wallis test (adjusted for ties) in the same BMI group; the *superscript letters* refer to the results of the Dunn's multiple comparison test; P^1 refers to the results of the Kruskal-Wallis test (adjusted for ties) in the same age group.

[a] $P < 0.01$ *vs.* adults; [b] $P < 0.01$ *vs.* elderly; [c] $P < 0.05$ *vs.* elderly.

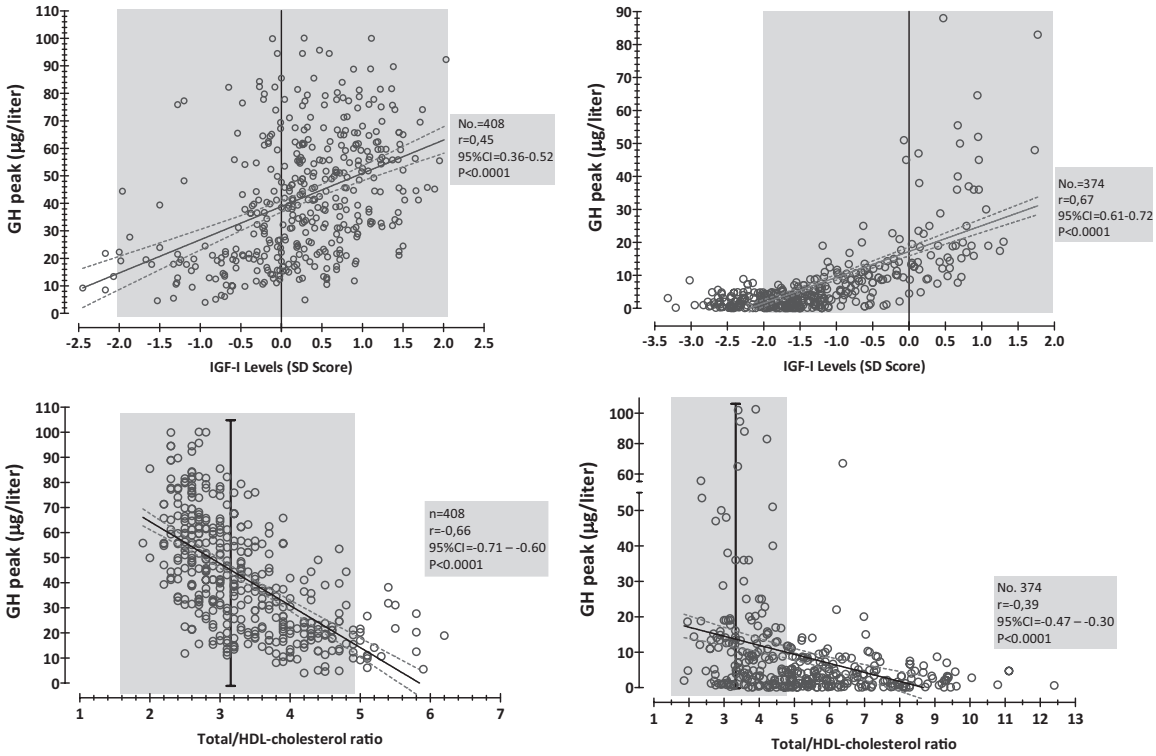

FIG. 1. Correlation analysis between GH peak after GHRH+ARG and IGF-I levels (*top panels*) measured as SDS or total to HDL cholesterol ratio (*bottom panels*) in the 408 controls (*left panels*) and the 374 hypopituitary patients (*right panels*). The *gray areas* show the mean ± 2 SD of control subjects.

jcem.endojournals.org). Of the 115 patients with isolated GHD or one more deficit, 37 of 40 patients with an IGF-I SDS of −1.00 or less had GHD (92.5%) compared with 33 of 75 (44.0%) with an IGF-I SDS greater than −1.00 ($P < 0.0001$). Of the 259 patients with two or more deficits, only one of 207 with an IGF-I SDS of −1.00 or less did not have a severe GHD (99.5%) compared with 20 of 52 patients with an IGF-I SDS greater than −1.00 ($P < 0.0001$).

Cutoff GH levels according to age and BMI by ROC

The analysis of GH cutoff after GHRH+ARG by ROC confirmed a higher GH peak in the young [19.1 μg/liter; sensitivity 83.1% (71.7–91.2), specificity 90.3% (83.7–94.9)] than in the adults [14.4 μg/liter; sensitivity 88.1% (83.4–91.8), specificity 90.3% (84.4–94.4)] and the elderly [9.7 μg/liter; sensitivity 91.8% (80.4–97.7), specificity 86.5% (78.4–92.4)] and in the lean [25.0 μg/liter; sensitivity 91.6% (86.1–95.4), specificity 92.4% (87.1–96.0)] than the overweight [14.4 μg/liter; sensitivity 89.9% (83.7–94.4), specificity 95.7% (90.3–98.6)] and the obese [6.2 μg/liter; sensitivity 90.0% (81.2–95.6), specificity 94.4% (88.3–97.9)]. The results of ROC analysis in the subjects grouped according to both age and BMI is shown Fig. 2. The results of the ROC analysis were in line with the results of the third percentile or slightly higher, except for the elderly obese group, in which the GH peak cutoff was 4.0 μg/liter, corresponding to the first percentile of control population. If the GH cutoff derived by ROC was considered to diagnose GHD, the prevalence increased from 286 to 337 of 374 (90.1%). The major increase in the prevalence of GHD was in the 26- to 65-yr-old patients in different BMI classes (Table 3).

The best IGF-I SDS by ROC to distinguish between GHD and non-GHD (according to the percentiles cutoff found in the current study) was −1.20 or less [sensitivity 80.5% (75.4–85.0), specificity 94.6% (87.8–98.2)]. All GHD patients had an IGF-I SDS of −1.50 or less.

Discussion

Worldwide, there is ongoing reappraisal of diagnostic tests to improve the diagnostic accuracy of GHD so that GH replacement could be used more appropriately to optimize cost/effectiveness of the treatment. This is the first study to propose a classification of GH cutoffs with a widely used and well-validated test, such as the GHRH+ARG, in a large series of patients with overt or suspected total or partial hypopituitarism, based on results obtained in a large group of healthy controls grouped according to both age and BMI. Previous studies proposed GH cutoff after the GHRH+ARG test only according to age (18, 21) or BMI (19, 20). Even taking into account that the GH assay changed during the study period, and thus, the GH cutoffs as crude

TABLE 3. GH peak after GHRH+ARG in the 366 hypopituitary patients grouped according with age and BMI. Data are shown as Mean ± SD and 95%CI in parentheses

	Young (15–25 yr)					Adults (26–65 yr)					Elderly (>65 yr)				
			Diagnosis of GHD					Diagnosis of GHD				Diagnosis of GHD			
	n	Peak GH (μg/liter)	1	2	3	n	Peak GH (μg/liter)	1	2	3	n	Peak GH (μg/liter)	1	2	3	P
Lean (L)	34	19.3 ± 36.8 (6.4–32.1)	25	26	26	113	9.9 ± 16.7 (6.8–13.1)	82	86	104	7	6.9 ± 7.5 (−0.05–13.8)	5	5	8	0.089
Overweight (OW)	21	8.8 ± 13.2 (2.8–14.8)	15	18	18	93	8.2 ± 12.7 (5.6–10.8)	63	59	85	25	5.4 ± 8.5 (1.9–8.9)	23	20	23	0.55
Obese (OB)	10	3.3 ± 4.4 (0.2–6.4)	9	9	10	54	3.3 ± 5.0 (1.9–4.7)	49	46	50	17	3.4 ± 4.2 (1.2–5.6)	14	13	13	0.99
	65		49	53	54	260		187	185	239	49		43	38	44	
P^1		0.076					<0.0001					0.33				
		L *vs.* OW 0.27					L *vs.* OW 0.67					L *vs.* OW 0.45				
		L *vs.* OB 0.021					L *vs.* OB 0.0002					L *vs.* OB 0.28				
		OW *vs.* OB 0.33					OW *vs.* OB <0.0001					OW *vs.* OB 0.23				

P refers to the results of the Kruskal-Wallis test (adjusted for ties) in the same BMI group; P^1 refers to the results of the Kruskal-Wallis test (adjusted for ties) in the same age group. GHD 1, Number of cases with GHD according to the old cutoff of 9 μg/liter (28); GHD 2, number of cases with GHD according to the GH cutoff reported in Table 2; GHD 3, number of cases with GHD according to the GH cutoff by ROC analysis reported in Fig. 2.

numbers should be considered cautiously, our data suggest that the GH threshold to perform an accurate diagnosis of GHD should be in line with individual patients' age and BMI. Even if we found that use of specific diagnostic cutoff by age and BMI minimally changed the prevalence of GHD in the current unselected population of patients with overt or suspected total or partial hypopituitarism, it reduced misdiagnosis in obese, elderly, and young subjects, compared with the old cutoff of 9.1 μg/liter, proposed by Aimaretti *et al.* (12) more than a decade ago. Importantly, a level of IGF-I SDS of −1.00 or less was diagnostic of GHD in 99.5% of patients with two or more pituitary deficits. An IGF-I SDS of −1.50 or less had 100% specificity independently from the number of pituitary deficits. These data could be of help in reducing the demand for GH testing with GHRH+ARG in patients with organic hypopituitarism to improve pharmacoeconomics of the diagnosis of GHD.

A recent consensus statement (26) revised the former one of 1998 (16) by stating that GHRH+ARG test is well validated in adults, being a successful alternative of insulin tolerance test because this latter test can be contraindicated in patients with ischemic heart disease or seizures and in the elderly. Additionally, the consensus statement also reported that one stimulation test is sufficient to diagnose GHD, and in patients with three or more pituitary hormone deficiencies and IGF-I level below the reference range, the likelihood of GHD is greater than 97%; therefore, a GH stimulation test is not required (26).

It is widely accepted that the diagnosis of any disease as well as relative therapeutic decisions should be based on well-defined criteria, ideally based on evidence of best practice reflecting outcome of international efforts to establish guidelines and consensus documents. Then for clinical purposes, a good test should discriminate between patients and controls with a higher than 90% accuracy, should be cheap and not laborious, and should be well tolerated by the patients. The GHRH+ARG test has been well validated in the last 20 yr to diagnose GHD; has excellent tolerability; is not laborious; has no need for a supervision of the patient during the test as during the ITT; and has been reported to correlate with patients' conditions in terms of lipid profile, bone density, and left ventricular performance (13–15). It should be noted that great variability in GH results from different assays has been reported from several countries by different laboratories participating in national external quality assessments (32). Another caveat to consider in the analysis of validity of the GHRH+ARG test is the stimulation both of the hypothalamus and the pituitary so that GHD due to hypothalamic disease may be missed. In fact, ITT showed a greater sensitivity and specificity within the first 5 yr after irradiation (33).

Studies in both the adult and pediatric literature support the concept of a continuum of peak GH responses to

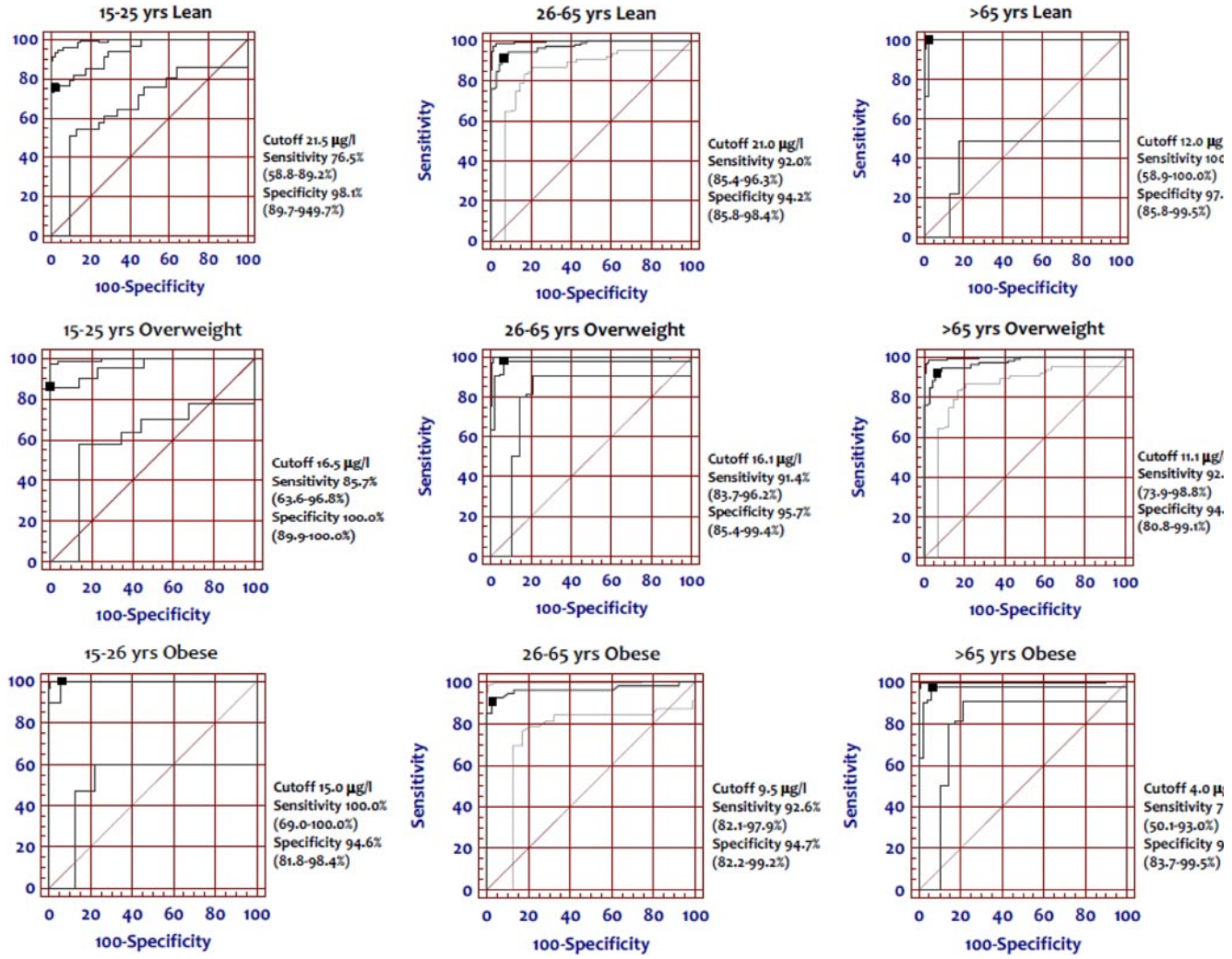

FIG. 2. Cutoff of GH peak after GHRH+ARG to distinguish between controls and hypopituitary patients according to ROC analysis.

standard GH stimulation tests between a normal peak GH response and severe GHD (11, 34, 35). In childhood, the peak GH of less than 5–7 μg/liter to ITT was validated against the height velocity both before and after GH therapy (11, 36). In the adult there is no specific end point equivalent to height velocity in children, which enables validation of our current biochemical definition of severe GHD. GHD in the adult is therefore defined biochemically as different GH peaks according to a stimulation test, age, and BMI of the patients (9, 10, 16–21, 26).

The results of the current study demonstrated that use of this test was helpful in diagnosis GHD in the setting of patients with organic hypopituitarism: only in a minority of cases (19 patients; 5.1%), the old cutoff of 9.1 μg/liter was not sufficiently sensitive to diagnose GHD, and this occurred mainly in obese patients, who need lower cutoff values, and the young lean, who need higher cutoff values. Additionally, we found that a level of IGF-I SDS of −1.00 or less diagnosed GHD with no need of a GH testing in 99.5% of the patients with more than two deficits and in 92.5% of those with one deficit or isolated GHD, whereas an IGF-I SDS of −1.50 or less diagnosed GHD in 100% of cases independently from BMI and number of associated pituitary deficiencies.

As a corollary finding of the current study, we reported that the GH cutoff derived from ROC analysis resulted in higher GH cutoff values than the percentiles analysis so diagnosing GHD in a larger proportion of cases. In our opinion, the GH cutoff based on percentiles has the advantage not only to enable a stricter definition of GHD but also to permit an estimation of partial GH deficiency, which will not be possible using ROC analysis. As mentioned in some studies in children (37) and adults (31–40), partial GHD, also called GH insufficiency, is associated with abnormal growth velocity, body composition, insulin sensitivity, cardiovascular risk, and performance, which are in between the severe GHD and the normal GH secretion. Even if partial GHD in adults is not recognized as a clinical entity (26), further studies might support initial data showing clinical features in partial as well as severe GHD in humans.

Conclusion

The results of the current study reported on different GH cutoff values in patients with organic hypopituitarism according to BMI and age, being the former the strongest predictor of GH peak after GHRH+ARG. Except for 5.1% of cases who benefit from a stricter diagnosis of GHD applying the new GH cutoff values, the old cutoff values proposed by Aimaretti *et al.* (12) distinguished appropriately between GHD and controls in 95% of cases. An IGF-I SDS of −1.00 or less enabled a correct diagnosis of GHD in 95% of patients with two or more pituitary deficiencies and in 92.5% of those with one deficiency or isolated GHD. Overall, an IGF-I SDS of −1.20 or less distinguished severe GHD from non-GHD with high sensitivity and specificity, whereas an IGF-I SDS of −1.50 or less diagnosed severe GHD in 100% of patients. These data might help in redesigning GH cutoff after GHRH+ARG in 15- to 83-yr-old patients and also selecting those patients who can be excluded from GH testing on the basis of number of pituitary deficiencies and IGF-I levels.

Acknowledgments

Address all correspondence and requests for reprints to: Annamaria Colao, M.D., Ph.D., Department of Molecular and Clinical Endocrinology and Oncology, "Federico II" University of Naples, via S. Pansini 5, 80131 Naples, Italy. E-mail: colao@unina.it.

This work was partially supported by Grant 7492 from Regione Campania L.R. 41/94 1999 and Grant 2003069821 (2003) from the Italian Minister of Research and University in Rome.

Disclosure Summary: A.C. is recipient of unrestricted grants from Ferring, Ipsen, Merck-Serono, and Pfizer for research programs in hypopituitarism; received lectures fees from Ipsen, Merck-Serono, and Pfizer; and is member of the scientific boards of Merck-Serono. G.A. is recipient of unrestricted grants from Ferring, Merck-Serono, Novo-Nordisk, and Pfizer for research programs in pituitary diseases and received lectures fees from Merck-Serono, Ely Lilly, Pfizer, and GlaxoSmithKline. None of the other authors have conflict of interest to disclose.

References

1. **Jørgensen JO, Pedersen SA, Thuesen L, Jørgensen J, Ingemann-Hansen T, Skakkebaek NE, Christiansen JS** 1989 Beneficial effects of growth hormone treatment in GH-deficient adults. Lancet 1:1221–1225
2. **Salomon F, Cuneo RC, Hesp R, Sönksen PH** 1989 The effects of treatment with recombinant human growth hormone on body composition and metabolism in adults with growth hormone deficiency. N Engl J Med 321:1797–1803
3. **Rosén T, Bengtsson BA** 1990 Premature mortality due to cardiovascular disease in hypopituitarism. Lancet 336:285–288
4. **Bates AS, van't Hoff W, Jones PJ, Clayton RN** 1996 The effect of hypopituitarism on life expectancy. J Clin Endocrinol Metab 81:1169–1172
5. **Bülow B, Hagmar L, Mikoczy Z, Nordstroem CH, Erfurth EM** 1997 Increased cerebrovascular mortality in patients with hypopituitarism. Clin Endocrinol (Oxf) 46:75–81
6. **Tomlinson JW, Holden N, Hills RK, Wheatley K, Clayton RN, Bates AS, Sheppard MC, Stewart PM** 2001 Premature mortality in 1014 patients with hypopituitarism. Lancet 357:425–431
7. **Svensson J, Bengtsson BA, Rosén T, Odén A, Johannsson G** 2004 Malignant disease and cardiovascular morbidity in hypopituitary adults with or without growth hormone replacement therapy. J Clin Endocrinol Metab 89:3306–3312
8. **Stochholm K, Gravholt CH, Laursen T, Laurberg P, Andersen M, Kristensen LØ, Feldt-Rasmussen U, Christiansen JS, Frydenberg M, Green A** 2007 Mortality and GH deficiency: a nationwide study. Eur J Endocrinol 157:9–18
9. **Gasco V, Corneli G, Beccuti G, Prodam F, Rovere S, Bellone J, Grottoli S, Aimaretti G, Ghigo E** 2008 Retesting the childhood-onset GH-deficient patient. Eur J Endocrinol 159(Suppl 1):S45–S52
10. **Casanueva FF, Castro AI, Micic D, Kelestimur F, Dieguez C** 2009 New guidelines for the diagnosis of growth hormone deficiency in adults. Horm Res 71(Suppl 1):112–115
11. **Hindmarsh P, Smith PJ, Brook CG, Matthews DR** 1987 The relationship between height velocity and growth hormone secretion in short prepubertal children. Clin Endocrinol (Oxf) 27:581–591
12. **Aimaretti G, Corneli G, Razzore P, Bellone S, Baffoni C, Arvat E, Camanni F, Ghigo E** 1998 Comparison between insulin-induced hypoglycemia and growth hormone (GH)-releasing hormone + arginine as provocative tests for the diagnosis of GH deficiency in adults. J Clin Endocrinol Metab 83:1615–1618
13. **Colao A, Cerbone G, Pivonello R, Aimaretti G, Loche S, Di Somma C, Faggiano A, Corneli G, Ghigo E, Lombardi G** 1999 The growth hormone (GH) response to the arginine plus GH-releasing hormone test is correlated to the severity of lipid profile abnormalities in adult patients with GH deficiency. J Clin Endocrinol Metab 84:1277–1282
14. **Colao A, Di Somma C, Pivonello R, Loche S, Aimaretti G, Cerbone G, Faggiano A, Corneli G, Ghigo E, Lombardi G** 1999 Bone loss is correlated to the severity of growth hormone deficiency in adult patients with hypopituitarism. J Clin Endocrinol Metab 84:1919–1924
15. **Colao A, Di Somma C, Cuocolo A, Filippella M, Rota F, Acampa W, Savastano S, Salvatore M, Lombardi G** 2004 The severity of growth hormone deficiency correlates with the severity of cardiac impairment in 100 adult patients with hypopituitarism: an observational, case-control study. J Clin Endocrinol Metab 89:5998–6004
16. **Growth Hormone Research Society** 1998 Consensus guidelines for the diagnosis and treatment of adults with growth hormone deficiency: summary statement of the Growth Hormone Research Society Workshop on Adult Growth Hormone Deficiency. J Clin Endocrinol Metab 83:379–381
17. **Clayton PE, Cuneo RC, Juul A, Monson JP, Shalet SM, Tauber M, European Society of Paediatric Endocrinology** 2005 Consensus statement on the management of the GH-treated adolescent in the transition to adult care. Eur J Endocrinol 152:165–170
18. **Maghnie M, Aimaretti G, Bellone S, Bona G, Bellone J, Baldelli R, de Sanctis C, Gargantini L, Gastaldi R, Ghizzoni L, Secco A, Tinelli C, Ghigo E** 2005 Diagnosis of GH deficiency in the transition period: accuracy of insulin tolerance test and insulin-like growth factor-I measurement. Eur J Endocrinol 152:589–596
19. **Biller BM, Samuels MH, Zagar A, Cook DM, Arafah BM, Bonert V, Stavrou S, Kleinberg DL, Chipman JJ, Hartman ML** 2002 Sensitivity and specificity of six tests for the diagnosis of adult GH deficiency. J Clin Endocrinol Metab 87:2067–2079
20. **Corneli G, Di Somma C, Baldelli R, Rovere S, Gasco V, Croce CG, Grottoli S, Maccario M, Colao A, Lombardi G, Ghigo E, Camanni F, Aimaretti G** 2005 The cut-off limits of the GH response to GH-

releasing hormone-arginine test related to body mass index. Eur J Endocrinol 153:257–264
21. **Corneli G, Di Somma C, Prodam F, Bellone J, Bellone S, Gasco V, Baldelli R, Rovere S, Schneider HJ, Gargantini L, Gastaldi R, Ghizzoni L, Valle D, Salerno M, Colao A, Bona G, Ghigo E, Maghnie M, Aimaretti G** 2007 Cut-off limits of the GH response to GHRH plus arginine test and IGF-I levels for the diagnosis of GH deficiency in late adolescents and young adults. Eur J Endocrinol 157:701–708
22. **Abs R, Feldt-Rasmussen U, Mattsson AF, Monson JP, Bengtsson BA, Góth MI, Wilton P, Koltowska-Häggström M** 2006 Determinants of cardiovascular risk in 2589 hypopituitary GH-deficient adults—a KIMS database analysis. Eur J Endocrinol 155:79–90
23. **Colao A, Di Somma C, Rota F, Di Maio S, Salerno M, Klain A, Spiezia S, Lombardi G** 2005 Common carotid intima-media thickness in growth hormone (GH)-deficient adolescents: a prospective study after GH withdrawal and restarting GH replacement. J Clin Endocrinol Metab 90:2659–2665
24. **Colao A, Di Somma C, Rota F, Pivonello R, Savanelli MC, Spiezia S, Lombardi G** 2005 Short-term effects of Growth Hormone (GH) treatment or deprivation on cardiovascular risk parameters and intima-media thickness at carotid arteries in patients with severe GH deficiency. J Clin Endocrinol Metab 90:2056–2062
25. **Colao A, Di Somma C, Cuocolo A, Spinelli L, Acampa W, Spiezia S, Rota F, Savanelli MC, Lombardi G** 2005 Does a gender-related effect of growth hormone (GH) replacement exist on cardiovascular risk factors, cardiac morphology, and performance and atherosclerosis? Results of a two-year open, prospective study in young adult men and women with severe GH deficiency. J Clin Endocrinol Metab 90:5146–5155
26. **Ho KKY, on behalf of the 2007 GH Deficiency Consensus Workshop Participants** 2007 Consensus guidelines for the diagnosis and treatment of adults with GH deficiency II: a statement of the GH Research Society in association with the European Society for Pediatric Endocrinology, Lawson Wilkins Society, European Society of Endocrinology, Japan Endocrine Society, and Endocrine Society of Australia. Eur J Endocrinol 157:695–700
27. **Grundy SM, Brewer Jr HB, Cleeman JI, Smith Jr SC, Lenfant C** 2004 Definition of metabolic syndrome: report of the National Heart, Lung, and Blood Institute/American Heart Association conference on scientific issues related to definition. Circulation 109:433–438
28. **Ghigo E, Aimaretti G, Gianotti L, Bellone J, Arvat E, Camanni F** 1996 New approach to the diagnosis of growth hormone deficiency in adults. Eur J Endocrinol 134:352–356
29. **Valetto MR, Bellone J, Baffoni C, Savio P, Aimaretti G, Gianotti L, Arvat E, Camanni F, Ghigo E** 1996 Reproducibility of the growth hormone response to stimulation with growth hormone-releasing hormone plus arginine during lifespan. Eur J Endocrinol 135:568–572
30. **Castelli WP** 1996 Lipid, risk factors and ischaemic heart disease. Atherosclerosis 124(Suppl):S1–S9
31. **Colao A, Di Somma C, Cascella T, Pivonello R, Vitale G, Grasso LF, Lombardi G, Savastano S** 2008 Relationships between serum IGF1 levels, blood pressure, and glucose tolerance: an observational, exploratory study in 404 subjects. Eur J Endocrinol 159:389–397
32. **Bidlingmaier M** 2008 Problems with GH assays and strategies toward standardization. Eur J Endocrinol 159:S41–S44
33. **Darzy KH, Aimaretti G, Wieringa G, Gattamaneni HR, Ghigo E, Shalet SM** 2003 The usefulness of the combined growth hormone (GH)-releasing hormone and arginine stimulation test in the diagnosis of radiation-induced GH deficiency is dependent on the post-irradiation time interval. J Clin Endocrinol Metab 88:95–102
34. **Toogood AA, Beardwell CG, Shalet SM** 1994 The severity of growth hormone deficiency in adults with pituitary disease is related to the degree of hypopituitarism. Clin Endocrinol (Oxf) 41:511–516
35. **Brennan BM, Rahim A, Mackie EM, Eden OB, Shalet SM** 1998 Growth hormone status in adults treated for acute lymphoblastic leukaemia in childhood. Clin Endocrinol (Oxf) 48:777–783
36. **Darendeliler F, Hindmarsh PC, Brook CG** 1990 Dose-response curves for treatment with biosynthetic human growth hormone. J Endocrinol 125:311–316
37. **Tauber M, Jouret B, Cartault A, Lounis N, Gayrard M, Marcouyeux C, Pienkowski C, Oliver I, Moulin P, Otal P, Joffre F, Arnaud C, Rochiccioli P** 2003 Adolescents with partial growth hormone (GH) deficiency develop alterations of body composition after GH discontinuation and require follow-up. J Clin Endocrinol Metab 88:5101–5106
38. **Murray RD, Adams JE, Shalet SM** 2004 Adults with partial growth hormone deficiency have an adverse body composition. J Clin Endocrinol Metab 89:1586–1591
39. **Murray RD** 2005 The phenotype of adults with partial growth hormone deficiency. Horm Res 64(Suppl 2):12–17
40. **Colao A, Di Somma C, Spiezia S, Rota F, Pivonello R, Savastano S, Lombardi G** 2006 The natural history of partial growth hormone deficiency in adults: a prospective study on the cardiovascular risk and atherosclerosis. J Clin Endocrinol Metab 91:2191–2200