

# The Severity of Growth Hormone Deficiency Correlates with the Severity of Cardiac Impairment in 100 Adult Patients with Hypopituitarism: An Observational, Case-Control Study

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In 100 patients with hypopituitarism and 80 sex- and age-matched healthy subjects, we correlated the severity of cardiac impairment to the severity of GH deficiency (GHD). By the GH peak after arginine plus GHRH test (normal > 16.5  $\mu\text{g/liter}$ ), the patients were classified as severe GHD (n = 56), partial GHD (n = 27), and non-GHD (n = 17).

Compared with controls, decreased left ventricular ejection fraction at rest was found only in severe GHD patients ( $55.0 \pm 8.8$  vs.  $63.4 \pm 4.5\%$ ,  $P < 0.001$ ); decreased left ventricular ejection fraction response on effort in severe ( $-4.6 \pm 17.4$  vs.  $15.2 \pm 9.1\%$ ,  $P < 0.001$ ) and partial GHD patients ( $3.6 \pm 6.6$  vs.  $14.6 \pm 8.3\%$ ,  $P < 0.001$ ); decreased diastolic filling at rest in severe ( $2.53 \pm 0.68$  vs.  $3.01 \pm 0.48$  end-diastolic volume per

second,  $P < 0.001$ ) and partial GHD ( $2.61 \pm 0.45$  vs.  $2.89 \pm 0.54$  end-diastolic volume per second,  $P = 0.004$ ) patients; and decreased exercise duration and capacity in all the patient groups. A normal systolic performance on effort was found in 21.4% of severe GHD, 55.6% of partial GHD, all non-GHD, and 93.7% of controls. A normal diastolic filling at rest was found in 57.1% of severe GHD, 74.1% of partial GHD, 76.5% of non-GHD, and 90% of controls.

In conclusion, cardiac performance is correlated with the GH status because significant impairment was found in patients with severe and partial GHD but not in non-GHD hypopituitary patients. (*J Clin Endocrinol Metab* 89: 5998–6004, 2004)

ADULTS WITH HYPOPITUITARISM have a reduced life expectancy with a higher than expected risk of death for cardio- and cerebrovascular disease, compared with healthy controls (1–4). Although a cluster of different factors, *i.e.* glucocorticoid and thyroid hormones overreplacement and/or gonadal steroids underreplacement, can potentially contribute to the increased cardiovascular mortality, a direct effect of GH deficiency (GHD) on the heart is highly reliable (5, 6). GHD can negatively influence cardiovascular function directly by impairing cardiac filling, performance, and contractility and indirectly by inducing hypercoagulability, abdominal obesity, insulin resistance, unfavorable lipid profile, impaired muscle performance, reduced pulmonary capacity, endothelial dysfunction, and atherosclerosis (5–9).

By echocardiography, in GHD patients with childhood-onset disease, reduced cardiac mass with an impaired systolic function was found in some but not all studies (6). By equilibrium radionuclide angiography, which enables a

more accurate estimation of cardiac performance on effort, we found an impaired left ventricular (LV) ejection fraction (LVEF), considered a main parameter of systolic function in the vast majority of GHD adults (10). Interestingly, reduced cardiac size was a more common finding in young patients with childhood-onset GHD (11, 12) and in children (13, 14) than in the adults, whereas impaired cardiac performance was not correlated with patients' age and was found in young (10), middle-aged (10), and elderly (15) patients. To further confirm a direct role of GHD in determining cardiac dysfunction, GH replacement was shown by us (14, 16–19) and others (11, 13, 20–23) to improve LVEF. Of note, adult GHD patients not receiving GH replacement were reported to have a further impairment of cardiac performance (18) and increased intima-media thickness at common carotid arteries (22). Furthermore, in severe GHD adolescents, withdrawal from GH replacement was followed by decreased cardiac size and reduced cardiac performance that were promptly reversed by GH treatment reinstatement (19).

We did previously show that the severity of GHD, measured as peak GH after the combined GHRH plus arginine (ARG) test, was correlated with the degree of abnormality of lipid profile (24) and bone loss (25). Of particular interest was the finding that patients with a GH peak after GHRH+ARG test between the first and the third percentile of normal according to Aimaretti *et al.* (26) and so diagnosed as having partial GHD showed abnormalities in lipid profile (24), body

Abbreviations: ARG, Arginine; BMI, body mass index; CV, coefficient of variation; DBP, diastolic blood pressure; EDV, end-diastolic volume; GHD, GH deficiency; IRMA, immunoradiometric assay; ITT, insulin tolerance test; LV, left ventricular; LVEF, LV ejection fraction;  $\Delta\text{LVEF}$ , change of LVEF; PFR, peak filling rate; SBP, systolic blood pressure.

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composition (24), and bone density (25) in between the patients with severe GHD and those without GHD. Our previous findings have been recently confirmed by Murray *et al.* (27) using the insulin tolerance test: these authors showed that adults with partial GHD (GH peak after insulin tolerance test between 3–7  $\mu\text{g}/\text{liter}$ ) have abnormal body composition similar to GHD and that the degree of this abnormality lied between that of healthy subjects and GHD adults and correlated with the IGF-I level.

To investigate whether the severity of GHD is also correlated with the degree of cardiac impairment and whether patients with partial GHD or even with some degree of pituitary dysfunction without GHD have alterations of cardiac performance, we designed this observational, case-control study. Cardiac performance, at rest and at peak exercise, was evaluated by equilibrium radionuclide angiography in 100 hypopituitary patients, each of them matched for gender and age with a healthy control. We considered as primary end point systolic performance, measured as LVEF at rest and at peak exercise and as secondary end points diastolic filling, measured as peak filling rate (PFR); hemodynamics, measured as heart rate; systolic (SBP) and diastolic blood pressure (DBP); and exercise performance, measured as duration and capacity of maximal physical exercise at cycloergometer. To avoid interference by obesity in this analysis, patients with body mass index (BMI) above 30  $\text{kg}/\text{m}^2$  were excluded.

## Patients and Methods

### Patients

One hundred nonobese hypopituitary patients (44 men, 56 women, aged 18–75 yr) diagnosed at the Department of Molecular and Clinical Endocrinology and Oncology of the University “Federico II” of Naples were included in this study. Exclusion criteria were: 1) present or previous concomitant diseases affecting cardiac function, such as chronically uncontrolled diabetes mellitus, coronary artery diseases, longstanding hypertension, or hyperthyroidism ( $n = 22$ ); 2) abnormal renal and/or hepatic function ( $n = 1$ ); and 3) BMI above 30  $\text{kg}/\text{m}^2$  ( $n = 19$ ). All patients had been previously operated on by transsphenoidal and/or transcranial route for nonfunctioning pituitary adenoma, meningioma, or craniopharyngioma. Sixteen patients had also been irradiated 3–7 yr before receiving testing for GHD. Before entering the study, the patients had undergone replacement therapy with L-thyroxine (50–100  $\mu\text{g}$  orally daily), cortisone acetate (25–37.5  $\text{mg}/\text{d}$ ), intranasal desmopressin (5–20  $\mu\text{g}/\text{d}$ ), testosterone-enanathate (250  $\text{mg}$  im monthly) in men, and oral and/or transdermal estrogens associated with progesterone in premenopausal females, as appropriate. Adequacy of hormone replacement therapy was periodically assessed by measuring serum free thyroid hormones, testosterone, urinary free cortisol and blood pressure, and serum  $\text{Na}^+$  and  $\text{K}^+$  measurements. At study entry, these hormonal parameters were in the normal range for age in all patients. None of the patients had ever received GH treatment. Sixty-eight patients were nonsmokers, whereas the remaining were mild smokers (less than 15 cigarettes/day).

### Controls

Eighty nonobese (BMI < 30  $\text{kg}/\text{m}^2$ ) healthy subjects were recruited among the medical and paramedical personnel of our department and their relatives and agreed to serve as controls: they were matched with the patients for age ( $\pm 1$  yr) and gender. Twenty healthy subjects served as case-control for two patients. The protocol of the study was approved by the Ethical Committee of the “Federico II” University of Naples, and all subjects gave their informed consent to the study. Fifty-five controls were nonsmokers (68.7%), whereas the remaining were mild smokers

(less than 15 cigarettes/day). Patients’ and controls’ profile, according to the diagnosis of GHD, at study entry is shown in Table 1.

### Study protocol

At study entry, in all cases we measured IGF-I levels and performed the GHRH+ARG test. ARG (arginine hydrochloride, Salf, Bergamo, Italy) was given at the dose of 0.5  $\text{g}/\text{kg}$ , up to a maximal dose of 30  $\text{g}$ , slowly infused from time 0 to 30 min, whereas GHRH (1–29, Geref, Serono, Rome, Italy) was given at the dose of 1  $\mu\text{g}/\text{kg}$  as iv bolus at time 0. Blood samples were taken every 15 min from –15 up to 90 min. Accordingly with previous findings in normal nonobese population (26), the GH response after ARG+GHRH was classified as follows: severe GHD when GH peak was less than 9  $\mu\text{g}/\text{liter}$ , partial GHD when GH peak was 9.1–16.5  $\mu\text{g}/\text{liter}$ , and normal GH response when GH peak was 16.5  $\mu\text{g}/\text{liter}$  or more. According to these criteria, 56 patients had severe GHD, 27 patients had partial GHD, and 17 patients with normal GH response were classified as non-GHD. All controls had a normal GH response to the test. Within 7–15 d from GH testing, all subjects were evaluated for cardiac performance by equilibrium radionuclide angiography. Heart rate and SBP and DBP were measured during the procedure by expert nuclear medicine physicians (W.A., A.Cu.), unaware whether studying a hypopituitary or a control subject.

### Equilibrium radionuclide angiography

*In vivo* labeling of red blood cells was performed with 555 MBq (15 mCi)  $^{99\text{m}}\text{Tc}$ . Radionuclide angiography was performed at rest and during dynamic physical exercise as previously described (15–18). A small field of view  $\gamma$ -camera (Starcam 300 A/M, General Electric, Milwaukee, WI) equipped with a low-energy, all-purpose collimator was used. Exercise studies were performed using a bicycle ergometer with a restraining harness to minimize patient motion under the camera. In all 180 subjects, exercise loads were increased by 25 W every 2 min until angina, limiting dyspnea, or fatigue developed. Heart rate, SBP, and DBP were monitored during exercise at each stage. None of the controls or hypopituitary patients developed high-grade ventricular arrhythmias necessitating termination of exercise; eight patients (8%) could not perform exercise due to very early muscular exhaustion. Radionuclide angiography studies were analyzed using a standard commercial software system (General Electric). LV regions of interest were automatically drawn for each frame, and a background region of interest was also computer delineated on the end systolic frame. After background correction, a LV time-activity curve was generated. Indexes of LV were derived by computer analysis of the background-corrected time-activity curve. LVEF was computed on the basis of relative on end-diastolic and end-systolic counts. Peak LV ejection and filling rates were also calculated after a Fourier expansion with four harmonics. PFR was computed as the maximum positive peak after end systole on the first derivative of LV time-activity curve and was computed in LV counts per second, normalized for number of counts at end diastole and expressed as end-diastolic volume (EDV) per second. Normal ranges were: PFR 2.5 or more EDV/sec; LVEF at rest 50% or more; exercise-induced changes of LVEF ( $\Delta\text{LVEF}$ ) an increase of 5% or more of LVEF at peak exercise, compared with resting values.

### Assays

Serum GH levels were measured by immunoradiometric assay (IRMA) using commercially available kits (HGH-CTK-IRMA, Sorin, Saluggia, Italy). The sensitivity of the assay was 0.2  $\mu\text{g}/\text{liter}$ . The intra- and interassay coefficients of variation (CVs) were 4.5 and 7.9%, respectively. Plasma IGF-I was measured by IRMA after ethanol extraction. The sensitivity of the assay was 0.8  $\mu\text{g}/\text{liter}$ . The normal IGF-I range in 20–40, 41–60, and over 60-yr-old subjects was 110–494, 100–300, and 78–260  $\mu\text{g}/\text{liter}$ , respectively. The intraassay CVs were 3.4, 3.0, and 1.5% for low, medium, and high point on the standard curve, respectively. The interassay CVs were 8.2, 1.5, and 3.7% for low, medium, and high point on the standard curve.

### Statistical analysis

The statistical analysis was performed by the SPSS package (SPSS Inc., Chicago, IL). Data are reported as mean  $\pm$  SD unless otherwise specified.

TABLE 1. Characteristics of patients and controls according to the GH response to GHRH + ARG test

	Severe GHD		Controls		Partial GHD		Controls		Non-GHD		Controls		P
	No. of patients	W/M	No. of patients	W/M	No. of patients	W/M	No. of patients	W/M	No. of patients	W/M	No. of patients	W/M	
Age (yr)	56	31/25	56	31/25	27	15/12	27	15/12	17	10/7	17	10/7	1
BMI (kg/m <sup>2</sup> )	43.4 ± 16.2		43.6 ± 16.3		41.7 ± 11.0		41.7 ± 11.1		33.7 ± 13.6		33.6 ± 13.6		0.98
Peak GH mean (±SD) (μg/liter)	25.9 ± 2.6		24.6 ± 6.4		25.3 ± 2.7		23.8 ± 3.0		24.1 ± 3.2		22.9 ± 2.8		0.001
IGF-I levels (μg/liter)	3.5 ± 3.0 <sup>a,b</sup>		53.0 ± 21.1		12.9 ± 1.9 <sup>a</sup>		43.9 ± 14.7		25.4 ± 9.3		55.0 ± 21.0		<0.001
z-score IGF-I	87.0 ± 40.4 <sup>a,b</sup>		236.4 ± 63.5		165.2 ± 20.9 <sup>a</sup>		232.7 ± 50.7		204.3 ± 48.2		274.2 ± 55.9		0.004
	-1.43 ± 0.72 <sup>a,b</sup>		0.52 ± 0.57		-0.36 ± 0.57		0.48 ± 0.67		-0.25 ± 0.58		0.50 ± 0.44		0.01

W, Women; M, men. The GH response after GHRH + ARG test was classified as follows: severe GHD when GH peak was  $\leq 9$  μg/liter, partial GHD when GH peak was 9.1–16.5 μg/liter, and non-GHD or normal response when GH peak was  $\geq 16.5$  μg/liter.

<sup>a</sup>  $P < 0.01$  vs. non-GHD; <sup>b</sup>  $P < 0.01$  vs. partial GHD patients.

Comparison between patients and controls was performed by the Mann-Whitney *U* test. Comparison among severe, partial, and non-GHD groups was performed by the Wilcoxon followed by the Dunn test. Significance was set at 5%. Categorical variables were compared using the Pearson's  $\chi^2$  test. Correlation coefficients were calculated by measuring the Pearson coefficient. The stepwise multiple linear regression was performed to evaluate the relative importance of age, BMI, peak GH after GHRH+ARG, IGF-I levels, and z-score of IGF-I on primary (LVEF at rest and at peak exercise) and secondary end points (PFR at rest and at peak exercise, SBP and DBP at rest and at peak exercise, heart rate at rest and at peak exercise, exercise duration and performance). In this analysis, we entered only those variables that had a  $P < 0.01$  in the univariate analysis.

## Results

Although BMI less than 30 kg/m<sup>2</sup> was an inclusion criterion for the study, the patients had a higher BMI than controls (Table 1). Serum IGF-I levels were lower than in controls in not only severe GHD but also partial GHD and non-GHD patients (Table 1). In both patients and controls, the GH peak to GHRH+ARG was correlated with age ( $r = -0.28$ ,  $P = 0.005$ ;  $r = -0.28$ ,  $P = 0.004$ ), BMI ( $r = -0.31$ ,  $P = 0.001$ ;  $r = -0.20$ ,  $P = 0.041$ ), and IGF-I levels ( $r = 0.84$ ,  $P < 0.0001$ ;  $r = 0.29$ ,  $P = 0.003$ ).

### Primary end point: GH peak after GHRH+ARG test vs. systolic performance

In both patients and controls, the GH peak to GHRH+ARG was correlated with LVEF at rest ( $r = 0.28$ ,  $P = 0.004$ ;  $r = 0.35$ ,  $P = 0.0004$ ) and at peak exercise ( $r = 0.57$ ,  $P < 0.0001$ ;  $r = 0.45$ ,  $P < 0.0001$ ) as well as with  $\Delta$ LVEF (Fig. 1). Patients with severe GHD ( $55.0 \pm 8.8$  vs.  $63.4 \pm 4.5\%$ ,  $P < 0.001$ ) and partial GHD ( $57.7 \pm 3.7$  vs.  $61.9 \pm 4.4\%$ ,  $P = 0.01$ ), but not non-GHD, had significantly lower LVEF at rest than controls (Fig. 2, top). Normal LVEF at rest was found in 44 severe GHD patients (78.6%) and all partial GHD and non-GHD patients and controls. LVEF response at peak exercise was significantly lower in severe ( $53.2 \pm 9.4$  vs.  $72.4 \pm 7.4\%$ ,  $P < 0.001$ ) and partial GHD patients ( $59.6 \pm 2.0$  vs.  $71.1 \pm 7.4$ ,  $P < 0.001$ ) than in controls and non-GHD (Fig. 2, middle). Non-GHD patients had a LVEF response at peak exercise similar to controls ( $70.9 \pm 5.2$  vs.  $73.5 \pm 5.9$ ,  $P = 0.18$ ). Eight patients with severe GHD could not perform the exercise test due to muscular exhaustion. A normal LVEF response at peak exercise was found in 12 severe GHD (21.4%), 15 partial GHD (55.6%), all non-GHD (100%), and 75 controls (93.7%,  $P = < 0.01$ ). So  $\Delta$ LVEF was significantly lower in severe ( $-4.6 \pm 17.4$  vs.  $15.2 \pm 9.1\%$ ,  $P < 0.001$ ) and partial GHD ( $3.6 \pm 6.6$  vs.  $14.6 \pm 8.3\%$ ,  $P < 0.001$ ) but not in non-GHD patients ( $16.4 \pm 6.8$  vs.  $17.0 \pm 10.8\%$ ,  $P = 0.53$ ) than in controls.  $\Delta$ LVEF in severe and partial GHD patients was significantly lower than in non-GHD patients ( $P < 0.001$ ; Fig. 2, bottom).

### Secondary end points: GH peak after GHRH+ARG test vs. diastolic filling, hemodynamics, and exercise performance

**Diastolic filling.** In both the patients and controls, the GH peak to GHRH+ARG was correlated with PFR at rest ( $r = 0.36$ ,  $P = 0.0002$  and  $r = 0.21$ ,  $P = 0.031$ ) and at peak exercise ( $r = 0.39$ ,  $P = 0.0001$  and  $r = 0.47$ ,  $P < 0.0001$ ). Normal PFR at rest was found in 32 severe GHD (57.1%), 20 partial GHD (74.1%),

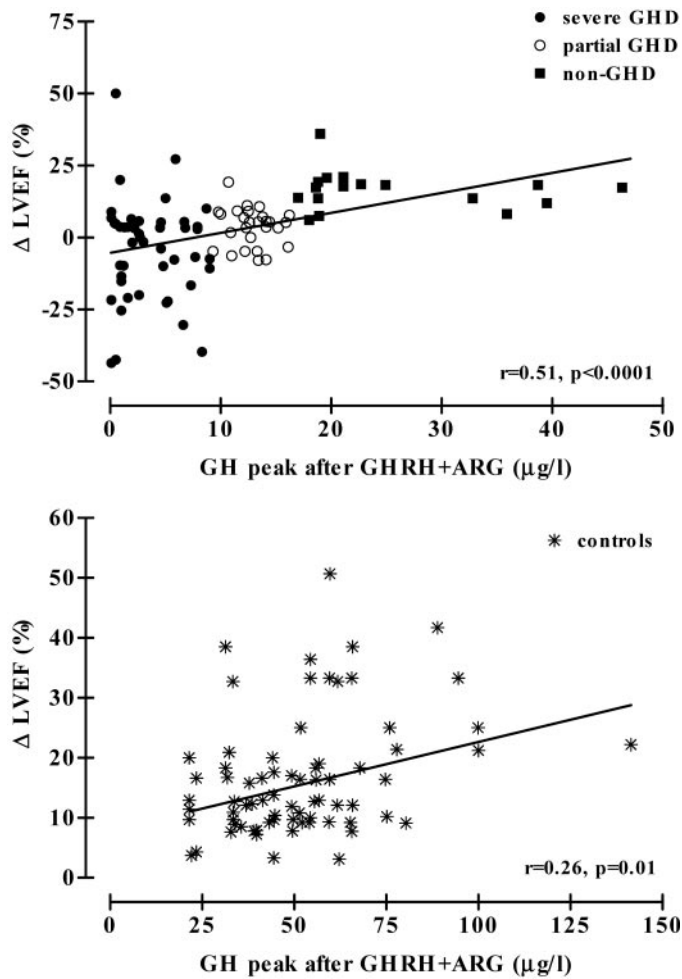


FIG. 1. Results of correlation analysis by calculating the Spearman's coefficient between GH peak after GHRH+ARG test and exercise-induced  $\Delta$ LVEF in 100 patients with hypopituitarism, grouped on the basis of the response to the text (*top*), and in their sex- and age-matched controls (*bottom*).

and 13 non-GHD patients (76.5%) and 72 controls (90%,  $P < 0.0001$ ). PFR at rest and at peak exercise was significantly lower in severe GHD patients than in controls and non-GHD patients (Table 2). Individual values of PFR in different groups are reported in Fig. 3.

**Hemodynamics.** The GH peak to GHRH+ARG was correlated in the patients with SBP at rest ( $r = -0.27$ ,  $P = 0.006$ ) and in the controls with SBP at rest and at peak exercise ( $r = -0.41$ ,  $P < 0.0001$  and  $r = -0.33$ ,  $P = 0.0008$ ) and DBP at rest and at peak exercise ( $r = -0.36$ ,  $P = 0.0002$  and  $r = -0.23$ ,  $P = 0.018$ ). Heart rate and SBP at peak exercise were lower in severe and partial GHD patients than in controls (Table 2).

**Exercise performance.** In both the patients and controls, the GH peak to GHRH+ARG was correlated with exercise duration ( $r = 0.46$ ,  $P < 0.0001$  and  $r = 0.52$ ,  $P < 0.0001$ ) and capacity ( $r = 0.41$ ,  $P < 0.0001$  and  $r = 0.20$ ,  $P = 0.043$ ). Exercise duration was significantly lower in all the patient groups than controls and was significantly lower in severe and partial GHD than in non-GHD (Table 2,  $P < 0.01$ ). Exercise capacity was significantly lower in all patient groups than

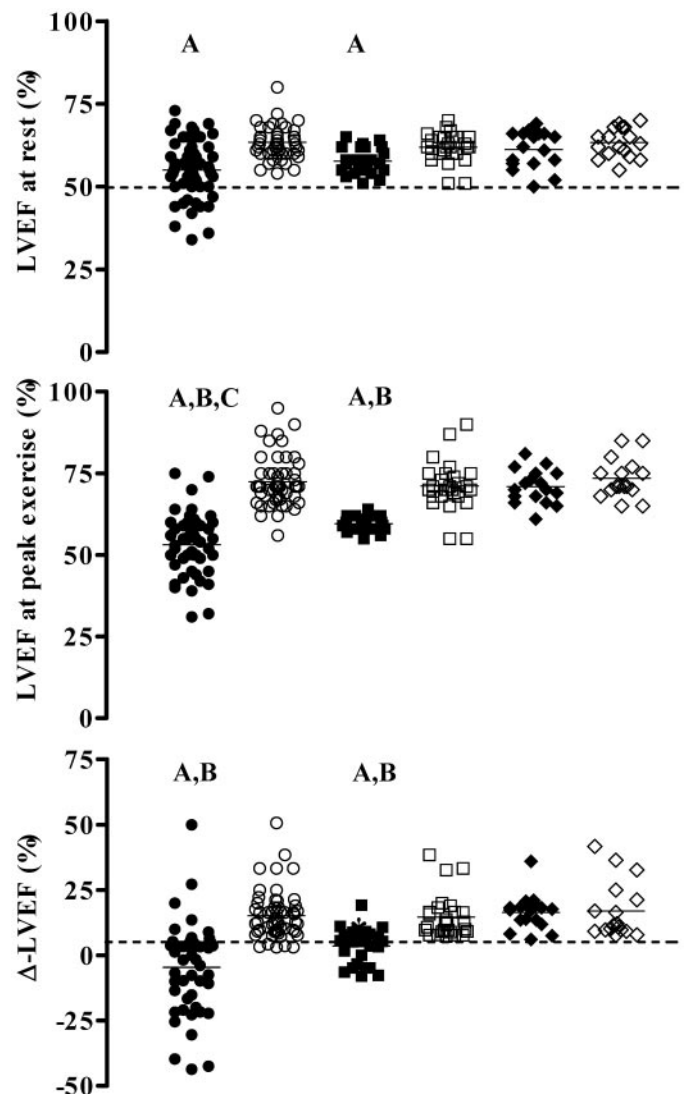


FIG. 2. Individual data of LVEF at rest (*top*), at peak exercise (*middle*), and exercise-induced  $\Delta$ LVEF (*bottom*) in the 56 patients with severe GHD (●) and their controls (○), the 27 patients with partial GHD (■) and their controls (□), and the 17 non-GHD patients (◆) and their controls (◇). The interrupted lines indicate the normal threshold of 50% for LVEF at rest and 5% of  $\Delta$ LVEF. A,  $P < 0.01$  vs. controls; B,  $P < 0.01$  vs. non-GHD; C,  $P < 0.01$  vs. partial GHD.

their controls and was significantly lower in severe GHD than in non-GHD (Table 2,  $P < 0.001$ ).

#### Results of the stepwise multiple linear regression (Table 3)

The GH peak after GHRH+ARG and/or IGF-I levels were the best predictors of LVEF at rest,  $\Delta$ LVEF, and exercise duration and capacity in both groups. In only the patients, IGF-I was the best predictor of PFR, whereas in the controls best predictor of PFR was BMI.

#### Discussion

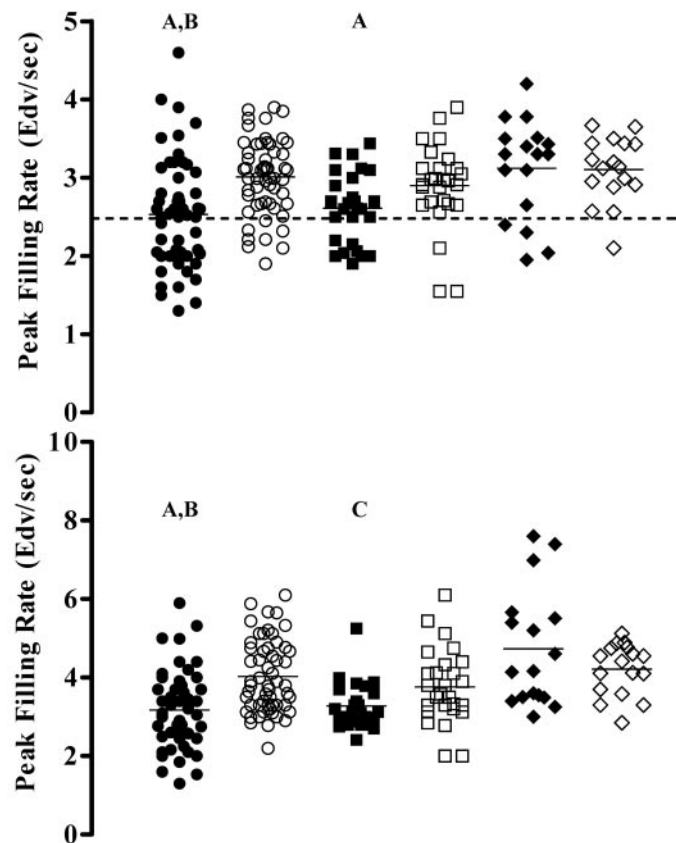
The results of this observational, case-control study show that the severity of GHD, evaluated on the basis of the GH response to GHRH+ARG test, correlates with the impair-

**TABLE 2.** Results of cardiac and exercise performance by equilibrium radionuclide angiography in patients and controls according to the response to GHRH + ARG test

	Severe GHD	Controls	<i>P</i>	Partial GHD	Controls	<i>P</i>	Non-GHD	Controls	<i>P</i>
PFR (EDV/sec)									
At rest	2.53 ± 0.68 <sup>a</sup>	3.01 ± 0.48	<0.001	2.61 ± 0.45 <sup>a</sup>	2.89 ± 0.54	0.004	3.12 ± 0.64	3.07 ± 0.29	0.27
At peak exercise	3.17 ± 1.01 <sup>a</sup>	4.02 ± 0.92	<0.001	3.28 ± 0.58 <sup>a</sup>	3.76 ± 0.94	0.04	4.73 ± 1.50	4.38 ± 1.04	0.62
Heart rate (bpm)									
At rest	77.3 ± 14.1	76.9 ± 6.8	0.79	76.4 ± 5.1	80.2 ± 6.0	0.98	80.8 ± 8.5	77.6 ± 6.3	0.44
At peak exercise	108.9 ± 22.2 <sup>a</sup>	125.2 ± 13.7	<0.001	108.4 ± 19.3 <sup>a</sup>	120.2 ± 19.6	0.005	127.3 ± 24.3	120.3 ± 17.3	0.08
SBP (mm Hg)									
At rest	120.6 ± 13.4	120.6 ± 11.6	0.88	118.7 ± 8.9	121.5 ± 13.5	0.64	113.5 ± 12.3	122.9 ± 10.8	0.08
At peak exercise	147.0 ± 20.6	154.5 ± 15.0	0.018	146.5 ± 12.9	156.5 ± 14.9	0.03	145.3 ± 15.3	154.1 ± 12.4	0.79
DBP (mm Hg)									
At rest	79.3 ± 8.8	79.3 ± 6.6	0.77	79.3 ± 5.5	78.7 ± 5.5	0.27	76.2 ± 7.6	78.5 ± 5.5	0.055
At peak exercise	95.5 ± 12.3	99.6 ± 12.1	0.19	94.6 ± 7.3	96.1 ± 7.9	0.3	92.6 ± 6.4	98.2 ± 10.4	0.059
Exercise duration (min)	6.1 ± 1.9 <sup>a,b</sup>	9.7 ± 1.2	<0.001	6.9 ± 1.1 <sup>a</sup>	9.2 ± 1.1	<0.001	8.2 ± 1.5	9.5 ± 1.3	<0.001
Exercise capacity (watts)	71.8 ± 28.6 <sup>a,b</sup>	101.7 ± 15.6	<0.001	81.3 ± 12.7 <sup>a</sup>	100.9 ± 16.2	0.004	100.0 ± 23.4	104.4 ± 13.2	0.017

Results at peak exercise refer to only 48 of 56 severe GHD patients, as the remaining patients could not perform exercise testing.

<sup>a</sup> *P* < 0.01 vs. non-GHD; <sup>b</sup> *P* < 0.01 vs. partial GHD patients.



**FIG. 3.** Individual data of LV PFR at rest (*top*), at peak exercise (*bottom*) in the 56 patients with severe GHD (●) and their controls (○), the 27 patients with partial GHD (■) and their controls (□), and the 17 non-GHD patients (◆) and their controls (◇). The interrupted lines indicate the normal threshold of 2.5 EDV/sec. A, *P* < 0.01 vs. controls; B, *P* < 0.01 vs. non-GHD; C, *P* < 0.05 vs. controls.

ment of systolic, diastolic, and exercise performance in patients with hypopituitarism. Interestingly, patients with deficiency of pituitary hormones other than GH, *i.e.* TSH (53%), ACTH (41%), FSH and LH (82%), treated with replacement therapy according to their deficiencies, have systolic performance, diastolic filling, and hemodynamics similar to con-

**TABLE 3.** Results of the stepwise multiple linear regression analysis of primary and second end-points of the study

		$\beta$ coefficient	<i>t</i>	<i>P</i>
GHD group best predictor(s)				
LVEF at rest	IGF-I	0.29	3.1	0.003
$\Delta$ -LVEF	GH peak	0.42	4.5	<0.0001
PFR	IGF-I	0.24	2.5	0.014
	Age	-0.24	-2.5	0.015
SBP at rest	Age	0.23	2.3	0.022
	GH peak	-0.2	-2.1	0.041
DBP at rest	Age	0.23	2.3	0.021
Heart rate at peak exercise	Age	-0.24	-2.4	0.019
Exercise duration	GH peak	0.43	4.5	<0.0001
Exercise capacity	GH peak	0.34	3.5	0.001
	Age	-0.21	-2.2	0.034
Control group best predictor(s)				
LVEF at rest	IGF-I	0.43	4.8	<0.0001
$\Delta$ -LVEF	GH peak	0.39	3.9	<0.0001
	IGF-I	0.22	2.2	0.03
PFR	BMI	0.54	7.0	<0.0001
	Age	-0.28	-3.7	<0.0001
SBP at rest	GH peak	-0.23	-2.3	0.021
SBP at peak exercise	BMI	0.34	3.3	0.001
	GH peak	-0.25	-2.4	0.02
	Age	0.23	2.2	0.031
DBP at rest	Age	0.43	4.7	<0.0001
Heart rate at peak exercise	IGF-I	0.22	2.2	0.03
Exercise duration	GH peak	0.51	6.0	<0.0001
	IGF-I	0.28	3.3	0.0002
Exercise capacity	GH peak	0.33	3.3	0.001
	IGF-I	0.28	2.8	0.007

trols. Conversely, even patients with a normal GH secretion and adequately replaced for their pituitary deficiency have a reduced duration and capacity of physical exercise, compared with age- and sex-matched controls. Moreover, patients with partial GHD, who generally do not undergo GH replacement therapy, also have impaired systolic performance, diastolic filling, hemodynamics, and exercise performance, compared with controls, even though not as relevant as the patients with severe GHD.

Several studies showed that patients with GHD have im-

paired cardiac performance (5, 6) both in resting conditions and on effort. We had previously demonstrated that adult patients with GHD have impaired systolic and diastolic performance (10, 15), partially reversed by 1-yr GH replacement at low doses (17) and further aggravated by 1 yr of GH deprivation (18). Slight but significant impairment of systolic and diastolic functions was also found in adolescent patients with severe GHD undergoing withdrawal of GH replacement at the achievement of final stature (19). However, although GH replacement improves cardiac performance (11, 14, 16–23), its final beneficial effect on cardiovascular and cerebrovascular mortality in GHD patients is still to be proven. In fact, despite the wide number of experimental evidences linking GHD to cardiac derangement (5, 6), the increased mortality of hypopituitary patients is considered to be due more likely to other negative factors, such as previous radiotherapy, glucocorticoid and thyroid hormone overreplacement, or gonadal steroid underreplacement (1–4). In particular, radiotherapy can have also direct effects in inducing cerebrovascular mortality, as suggested by a recent study in the cohort of patients with acromegaly (28). Radiotherapy had been performed several years before entering the study in 16 of the 100 patients: 14 of the 16 were classified as severe GHD and two as partial GHD. Gleeson *et al.* (29) recently reported that cranial irradiation during childhood for cancer does not inevitably cause severe GHD in adulthood and that these patients require retesting. Our patients were all diagnosed after the age of 16 yr, after their final height had been achieved, and were tested 3–7 yr after radiotherapy so that the diagnosis is likely to be appropriate. Of the two irradiated patients with partial GHD, one was a 20-yr-old woman who received radiotherapy 3.5 yr before entering the study, with a peak GH after GHRH+ARG of 10.5  $\mu\text{g}/\text{liter}$  and an IGF-I z-score of  $-1.2$ , and the other was a 33-yr-old man who received radiotherapy 4 yr before entering the study, with a peak GH after GHRH+ARG of 13.5  $\mu\text{g}/\text{liter}$  and an IGF-I z-score of  $-0.75$ . It should be mentioned, however, that the most recent study focusing on the mortality of hypopituitary patients (4) could not demonstrate any effect of GHD on mortality, probably due to the limited number of cases undergoing diagnostic testing for GHD. This is not surprising considering that the GHD syndrome in adult subjects has been described only in the past 15 yr.

The GHD syndrome is characterized by a cluster of detrimental factors for the cardiovascular system, *i.e.* high cholesterol levels, low high-density lipoprotein-cholesterol levels, high body fat mass, low muscle mass and performance, endothelial dysfunction, and precocious atherosclerosis (5–9). To diagnose GHD in adult patients, the insulin tolerance test (ITT) and GHRH+ARG test are considered gold standards (30). In particular, it is relevant to note that the first percentile of normal response to ITT gives a threshold value of 3  $\mu\text{g}/\text{liter}$ , whereas the identical threshold of the GHRH+ARG test is 9  $\mu\text{g}/\text{liter}$  (26). This broader range to establish GHD by the more potent GHRH+ARG test enables a more accurate estimation of the severity of GHD. In fact, by using the GHRH+ARG test, we demonstrated that the GH response correlated with the severity of lipid abnormalities (24), alterations of body composition (24), and bone loss (25). BMI has recently been shown to affect the GH response to the

GHRH+ARG test. In fact, Biller *et al.* (31) compared five different GH stimulation tests for diagnosing adult GHD in patients with pituitary diseases and healthy controls matched with the patients for age, sex, BMI, and estrogen use. They reported a GH peak cut-off to discriminate GHD patients and controls of 4.1  $\mu\text{g}/\text{liter}$ , so it was lower than that reported by Aimaretti *et al.* (26) of 9  $\mu\text{g}/\text{liter}$ . However, in the U.S. series, both patients and controls had a higher BMI than the patients we studied in the past (24, 25) as well as those included in the present study. To keep our old diagnostic criteria (24, 25), also considering the results of the U.S. series (31), in the current study, we excluded the patients with a BMI 30  $\text{kg}/\text{m}^2$  or more: this should have ruled out any effect of obesity in determining the alterations of cardiac and exercise performance. In line with Biller *et al.* (31), in our series the GH peak after GHRH+ARG was significantly correlated with BMI in both the patients and controls, although in these latter, the correlation was faint.

In this study, we show that GHD has clear-cut negative effects on systolic performance, diastolic filling, and ability to perform physical exercise. The severity of GHD, measured as GH peak to GHRH+ARG test, was correlated with the severity of systolic impairment, measured as LVEF on effort, and diastolic performance, measured as PFR. Besides, the GH peak to GHRH+ARG was also correlated with  $\Delta\text{LVEF}$  in our controls: this result was likely due to the fact that there is an important effect of age in determining a higher GH response and a higher  $\Delta\text{LVEF}$  in the young than middle-aged and elderly subjects. However, because the results were analyzed in an age-matched, case-control study, the effect of age was irrelevant in the observed difference between the patients and controls. Interestingly, the patients having other pituitary deficiencies but GH, thus classified as non-GHD, and receiving appropriate hormone replacement according to current clinical practice had a normal cardiac performance and an impaired exercise performance. These data, even if collected in a still small series of patients, indicate that the current replacement therapies with gonadal steroids, glucocorticoids, and thyroid hormones do not negatively affect cardiac performance. It is relevant to note that the patients with a GH peak to GHRH+ARG between 9 and 16.5  $\mu\text{g}/\text{liter}$ , the first and third percentile of normalcy according to Aimaretti *et al.* (26), had impaired systolic, diastolic, and exercise performance, compared with healthy controls, like the patients with severe GHD, but impairment was less relevant than these latter. Recently Murray *et al.* (27), using the ITT, have also shown that adults with partial GHD, by a GH peak after ITT between 3 and 7  $\mu\text{g}/\text{liter}$ , have abnormal body composition similar to the patients with severe GHD. These findings, which agree with our previous results (24, 25), suggest that patients with partial GHD should be carefully followed up and eventually retested to further investigate the clinical implications of reduced GH secretion.

In conclusion, GHD is characterized by an impairment of cardiac performance demonstrated by an abnormal response of the LVEF and an inadequate diastolic filling. The prevalence of systolic and diastolic derangement is higher in patients with severe GHD than in those with partial GHD. Patients with a normal GH response to the GHRH+ARG test have a normal cardiac performance. These results further substantiate the detrimental effect of GHD on the heart and

indicate that the GHRH+ARG test is a reliable method to investigate the GH deficiency in adults because its results correlate with clinical end points, such as cardiac and exercise performance.

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