Clinical Endocrinology (2001) **54**, 515–524

Increased arterial intima-media thickness by B-M mode echodoppler ultrasonography in acromegaly

Annamaria Colao*, Stefano Spiezia†, Gaetana Cerbone*, Rosario Pivonello*, Paolo Marzullo*, Diego Ferone*, Carolina Di Somma*, Angelo P. Assanti† and Gaetano Lombardi*

**Department of Molecular and Clinical Endocrinology and Oncology, 'Federico II' University of Naples and †Echo-guided Surgery Unit, 'S. Maria del Popolo degli Incurabili' Hospital, Naples, Italy*

(Received 5 October 1999; returned for revision 14 December 1999; finally revised 21 July 2000; accepted 8 September 2000)

Summary

BACKGROUND Patients with acromegaly have an increased morbidity and mortality for cardiovascular diseases. Despite the increasing evidence for the existence of a specific cardiomyopathy in acromegaly, the presence of vascular abnormalities has been never investigated.

OBJECTIVE To evaluate the cardiovascular risk and premature atherosclerosis in acromegaly.

SUBJECTS Forty-five patients with acromegaly and 30 sex- and age-matched healthy subjects were included in this study: 30 patients were studied at the diagnosis of acromegaly and were in active disease (GH 59·3 ± 10·2 mU/l, IGF-I 733 ± 57·6 μg/l) while 15 patients were studied after surgery and/or radiotherapy and were cured from the disease (GH 4·5 ± 0·7 mU/l, IGF-I 172·4 ± 16·9 μg/l).

METHODS Body mass index (BMI), systolic (SBP) and diastolic blood pressure (DBP), serum total, LDL- and HDL-cholesterol, triglycerides, and fibrinogen levels, prothrombin time (PT), activated partial thromboplastine time (APTT), glucose and insulin levels (fasting and after glucose load) were measured in all patients and controls. By echodoppler ultrasonography, blood systolic (SPV) and diastolic (DPV) peak velocity, and resistance index (RI) were measured at both common and internal carotid arteries where presence, size and location of atherosclerotic plaques were evaluated by B-Mode ultrasonography. Intima-media thickness (IMT) of both common carotids was measured by M-Mode ultrasonography.

RESULTS SBP, but not DBP, was significantly higher in patients with active disease than in cured patients and controls ($P = 0{\cdot}003$). Hypertension was found in nine (30%) patients with active disease, in two (13·3%) of those cured from acromegaly and in none of controls ($\chi^2 = 10{\cdot}81$, $P < 0{\cdot}004$). Fasting blood glucose levels were significantly higher both in patients with active disease and in those cured from the disease than in controls ($P < 0{\cdot}001$). Circulating insulin levels were significantly higher in patients with active disease than in cured patients and controls ($P < 0{\cdot}001$) and in cured patients than in controls ($P < 0{\cdot}001$). Glucose tolerance abnormalities were found in 13 (43·3%) patients with active disease, in four (26·6%) patients with inactive disease and in four controls (13·3%) ($\chi^2 = 6{\cdot}71$, $P = 0{\cdot}03$). Total blood cholesterol levels were similar in the three groups, LDL-cholesterol and triglycerides levels were significantly higher, whereas HDL-cholesterol levels were significantly lower both in patients with active disease and in those cured from the disease than in controls ($P < 0{\cdot}001$). Serum fibrinogen levels were significantly higher both in patients with active disease and in those cured from the disease than in controls ($P < 0{\cdot}001$). No difference was found in PT and APTT levels among the three groups.

At the level of right and left common carotid arteries, IMT was significantly higher both in patients with active disease and in those cured from the disease than in controls ($P < 0{\cdot}001$). Both right and left SPV, but not DPV, were significantly higher in patients with active disease than in those cured from the disease and in controls ($P < 0{\cdot}01$). Well defined carotid wall plaques were detected in two patients (6·6%) with active disease, in one patient cured from the disease (6·6%) and in two controls (6·6%). At the level of right and left internal carotid arteries, SPV, DPV and RI were similar among the three groups. Well defined carotid wall plaques were detected in three patients with active disease (10%), two patients cured from the disease (13·3%) and in one control (3·3%).

CONCLUSIONS A significant increase of IMT of both common carotid arteries was observed in patients

Correspondence: Dr Annamaria Colao, Department of Molecular and Clinical Endocrinology and Oncology, 'Federico II' University of Naples, Via Sergio Pansini 5, 80131, Naples, Italy, Fax: + 39 0817463668, E-mail: colao@unina.it

with active acromegaly, this was also found in those cured from acromegaly. However, the prevalence of well defined carotid plaques was not increased in both groups of patients with acromegaly as compared to controls. On this basis, heart more than vessels seems to be affected by chronic GH and IGF-I excess in acromegaly.

Atherosclerosis, cardiovascular and cerebrovascular diseases double the death rate in patients with acromegaly as compared to healthy population, mainly after the age of 45 years (Wright *et al*., 1970; Nabarro, 1987; Bengtsson *et al*., 1988; Orme *et al*., 1998). The poor prognosis of acromegalic patients is not only due to an increased frequency of cardiovascular disease, such as systemic arterial hypertension, and premature coronary artery disease, but also to the presence of a specific cardiomyopathy (Colao *et al*., 1997; Saccà *et al*., 1994; Lombardi *et al*., 1997; López-Velasco *et al*., 1997; Colao & Lombardi, 1998). Chronic GH and IGF-I excess has been demonstrated to cause a derangement of cardiomyocytes leading to abnormalities in cardiac muscle structure and function (Colao *et al*., 1999a; Lopez-Velasco *et al*., 1997; Frustaci *et al*., 1999), which were partially reversed by suppressing GH and IGF-I levels (Baldelli *et al*., 1999; Thuesen *et al*., 1989; Chanson *et al*., 1990; Pereira *et al*., 1991; Merola *et al*., 1993; Colao *et al*., 1999b; Hradec *et al*., 1999).

The harmful effect of GH/IGF-I excess on cardiac structure and function has been widely demonstrated by *in vivo* and *in vitro* studies while no data are available on the vascular consequences of acromegaly. Circulating IGF-I levels seem to play a relevant role on endothelial function as IGF-I was demonstrated to increase nitric oxide (NO) production at that level (Walsh *et al*., 1996). Furthermore, other independent risk factors such as high glucose, insulin, cholesterol, triglycerides and fibrinogen levels are considered as responsible for the increased cardiovascular risk in acromegaly (Melmed, 1990).

The current study was designed to investigate on the cardiovascular risk and vascular consequences of patients with acromegaly. At this purpose, a B- and M-Mode echodoppler ultrasonography study of both common and internal carotid arteries together with measurement of the main parameters associated with cardiovascular risk were performed in naive patients with acromegaly, in those cured from the disease and in healthy controls.

Subjects and methods

Patients

Forty-five patients with acromegaly (men 22, women 23, age 18–65 years) entered this open transversal study. Acromegaly was diagnosed in keeping with typical clinical features, high serum GH levels during a 8-h time course, not suppressible below 6 mU/l after 75 g oral glucose load and high plasma IGF-I levels for age (Colao *et al*., 1997). The cure from the disease was considered when safe hormone levels were achieved, i.e. fasting or glucose-suppressed GH levels were below 7·5 or 3 mU/l (equal to 2·5 and 1 μg/l), respectively, together with normal IGF-I levels for age (Clayton, 1997; Colao & Lombardi, 1998). Thirty out of 45 patients were studied at the diagnosis of acromegaly and were in active disease (GH 59·3 ± 10·2 mU/l, IGF-I 733 ± 57·6 μg/l) while 15 patients were cured from the disease (GH 4·5 ± 0·7 mU/l, IGF-I 172·4 ± 16·9 μg/l). Circulating GH and IGF-I (after ethanol extraction) levels were assayed by immunoassays using commercially available kits. In our laboratory the normal IGF-I range in adults aged 20–30, 31–40, 41–50, over 50-year-old subjects was 110–502, 100–494, 100–303, and 78–258 μg/l, respectively. The presumed duration of acromegaly was assessed by comparison of patients photographs taken during one-to three-decade span and by patient's interviews to date the onset of acral enlargement and it was assumed as the interval between the clinical onset and the time of treatment. In the present series of patients, disease duration ranged between 4 and 30 years. The cure from the disease occurred between 5 and 15 years before entering the study. Thirty healthy subjects, among the medical and paramedical personnel of our department, sex- and age-matched with the patients agreed to participate in this study and were used as controls. Exclusion criteria were familial or personal history of cardiovascular diseases and previous treatment with drugs known to interfere with glucose or lipid metabolism or influence blood pressure. Twenty-two patients (48·8%) and 15 controls (50%) were nonsmokers, two patients were ex-smokers, and all had a sedentary lifestyle. All patients and controls gave their informed consent to participate in this study and the study protocol was approved by the ethical committee of the Medical School of the University 'Federico II' of Naples. At study entry, two patients with active acromegaly were treated with ACE-inhibitors for moderate hypertension.

Study protocol

All patients and controls were submitted to a clinical, biochemical and vascular study.

Clinical study. Body mass index (BMI), systolic (SBP) and diastolic blood pressure (DBP) were evaluated by standard methods. A BMI between 25 and 30 was considered as index of overweight while >30 as index of obesity. Blood pressure was measured in the right arm, with the subjects in relaxed

Table 1 Clinical, biochemical and hormonal features of the 75 subjects included in the study

Parameters	Acromegalic patients		Controls	*P*
	Active disease ($n = 30$)	Cured ($n = 15$)	($n = 30$)	
Age [range/mean ± SEM] (years)	18–65 (38·5 ± 2·5)	25–60 (40·0 ± 2·9)	18–65 (38·5 ± 2·5)	
Men/women	17/13	5/10	15/15	
Body Mass Index (kg/m^2)	25·6 ± 1·2	22·8 ± 1·1	23·5 ± 0·5	0·1
Fasting GH levels (mU/l)	59·3 ± 10·2*	4·5 ± 0·7	3·0 ± 0·5	0·000
Plasma IGF-I levels (μg/l)	733·0 ± 57·6*	172·4 ± 16·9	248·4 ± 16·7	0·000
Systolic blood pressure (mmHg)	132·7 ± 2·9*	121·4 ± 4·9	117·7 ± 2·9	0·003
Diastolic blood pressure (mmHg)	87·9 ± 2·4	80·0 ± 3·2	80·7 ± 2·9	0·09
Fasting blood glucose levels (mmol/l)	5·9 ± 0·2	5·4 ± 0·5	4·7 ± 0·1†	0·000
Fasting serum insulin levels (pmol/l)	232·5 ± 15·8*	141·3 ± 24·4‡	75·3 ± 22·2	0·000
Serum triglycerides levels (mmol/l)	1·3 ± 0·1	1·7 ± 0·4‡	1·1 ± 0·05	0·03
Total blood cholesterol levels (mg/dl)	196·5 ± 7·5	189·7 ± 5·7	187·3 ± 5·1	0·5
LDL-cholesterol levels (mg/dl)	82·3 ± 4·4§	100·8 ± 4·9‡	87·4 ± 3·3	0·02
HDL-cholesterol levels (mg/dl)	39·4 ± 1·4	41·4 ± 2·4	60·3 ± 2·2†	0·000
Plasma fibrinogen levels (μmol/l)	12·35 ± 0·6	12·6 ± 0·6	5·8 ± 0·2†	0·000
Activated partial thromboplastin time (sec)	24·7 ± 0·5	24·1 ± 0·6	26·6 ± 1·1	0·1
Prothrombin time (%)	95·9 ± 1·5	93·8 ± 1·7	94·6 ± 1·7	0·7

*Significant *vs.* cured patients and healthy controls; †significant *vs.* active and cured patients; ‡significant *vs.* healthy controls; §significant *vs.* cured patients.

sitting position. The average of six measurements (three taken by each of two examiners) with a mercury sphygmomanometer was used. Hypertension was diagnosed when diastolic blood pressure (DBP) values were >90 mmHg and was graded as mild when between 91 and 104 mmHg, moderate when 105–114 mmHg, and severe when >115 mmHg, in line with WHO criteria (1996).

Metabolic study. Fasting glucose, triglycerides, total, LDL-and HDL-cholesterol, prothrombin time (PT), activated partial thromboplastine time (APTT), and fibrinogen levels were measured by standard procedures. Hypertriglyceridaemia was diagnosed when triglycerides levels were $>2{\cdot}8$ mmol/l (Consensus Conference, 1984) while hypercholesterolaemia was diagnosed when total cholesterol levels were >240 mg/dl (Expert panel 1988). Impaired glucose tolerance was diagnosed after an oral glucose tolerance test (75 g of glucose diluted in 250 ml of saline solution, measuring blood glucose every 30 minutes for 2 h). The diagnosis of diabetes mellitus or impaired glucose tolerance was performed according to the following criteria. Diabetes mellitus was diagnosed when fasting glucose was above 7 mmol/l at two consecutive measurements or when 2 h after the oGTT glucose was $\geq 11{\cdot}1$ mmol/l. Impaired glucose tolerance was diagnosed when glucose was between 7 and 11·1 mmol/l 2 h after the oGTT with an additional value $\geq 11{\cdot}1$ mmol/l between 0 and 2 h after glucose load (The DECODE study, 1999). Fasting and glucose load-stimulated insulin levels were measured in patients and controls to estimate insulin sensitivity. Hyperfibrinogenaemia was diagnosed when fibrinogen levels were >4 g/l.

Vascular study. Common and internal carotid arteries ultrasound imaging was carried out with a Vingmed Sound CMF 725 equipment (Horten, Norway) by means of a 7·5-MHz annular phased array transducer. Details on the technique were reported elsewhere (Colao *et al*., 1999c). Right and left carotid arteries were scanned longitudinally, 2·5 cm proximal and 1 cm distal to the bifurcation. When satisfactory B-mode imaging of common carotid artery wall was achieved, M-mode images were taken for several cardiac cycles to obtain the best quality measurements of intima-media thickness (IMT). Quantitative and semiquantitative indices were evaluated by echodoppler ultrasonography (US) placing the sample volume (set at 75% of lumen calibre) in the middle of the vessel lumen. All pictures were stored on magnetic media and analysed later. US imaging studies were performed by two operators (S.S., A.P.A.) blind in respect to the phase of disease activity in the patients. Each measurement was repeated thrice by the single investigator and the mean of the 6 evaluations taken into consideration. The variability in the intima-media thickness measurement for our instrument was 0·03 mm. Flow indices of both carotid were investigated by measuring blood systolic (SPV) and diastolic (DPV) peak velocities, and resistance (RI) index. The IMT was measured at the level of both common

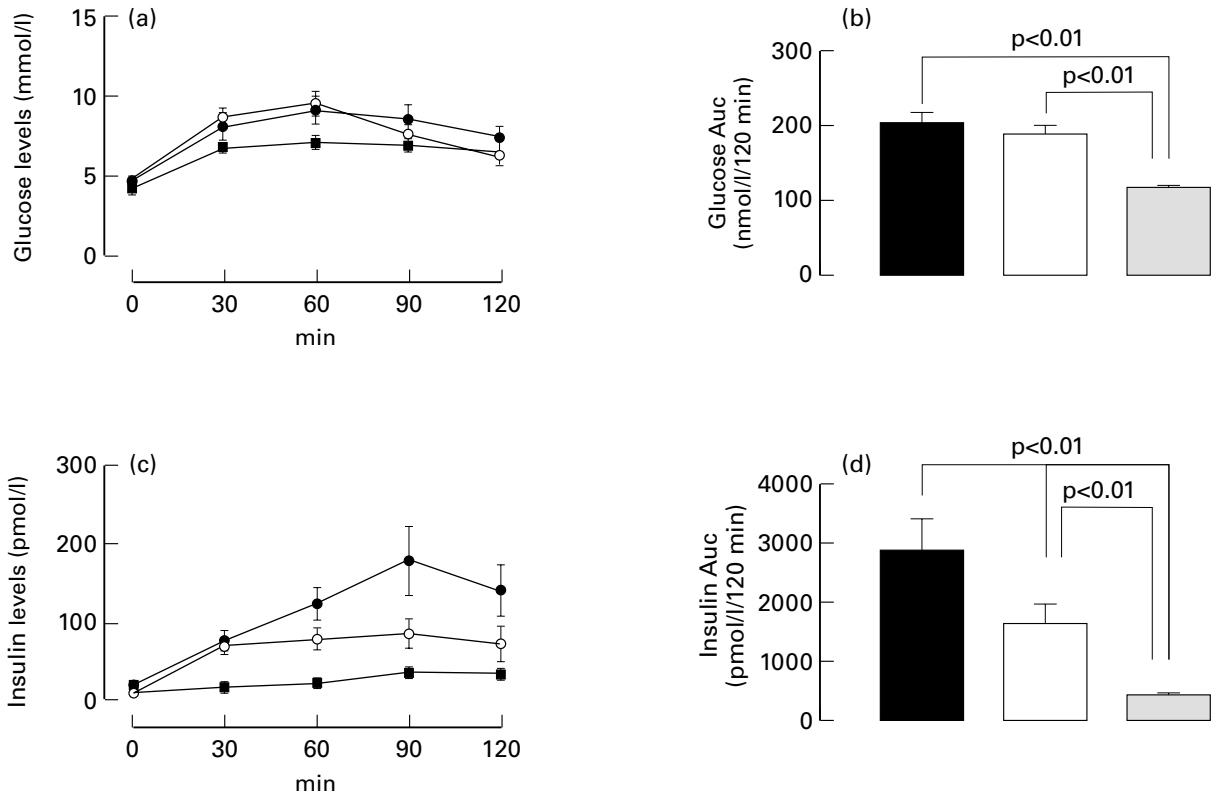

Fig. 1 (a) Blood glucose and (c) serum insulin response to oral glucose tolerance test in patients with active (●) and cured (○) acromegaly and controls (■). Data are shown as Mean ± SEM. The areas under the curves for (b) blood glucose and (d) serum insulin are shown as active (■) and cured (□) acromegaly and controls (▒).

carotid arteries and was considered normal when < 1 mm (Pignoli *et al*., 1986; Markussis *et al*., 1992; Gamble *et al*., 1994; Colao *et al*., 1999c). The ratio between the DPV of internal vs. common carotid artery was calculated to evaluate the presence of vascular obstructions distal to the measurement: a ratio between 1·5 and 2 was considered diagnostic of a mild (30%) stenosis, between 2·1 and 2·5 of a moderate (50%) stenosis; between 2·6 and 3 of a severe (75%) stenosis and above 3 of a very severe (90%) stenosis (Gamble *et al*., 1994; Pignoli *et al*., 1986). The presence, location and size of plaques were also evaluated at the level of common and internal carotid arteries. A type IV plaque featured by thickening of vascular wall and increased density of all US-evaluable layers without any haemodynamic alteration was defined as a well-defined plaque (Belcaro & Laurora, 1993).

Statistical analysis. The statistical analysis was performed by means of the SPSS Inc. (Cary, NC, USA) package. Data are reported as Mean ± SEM. Glucose and insulin response to glucose load was reported as peak and area under the curve (Auc) calculated by the trapezoid integration method. The statistical analysis was performed by ANOVA. The significance was set at 5%. Post-hoc analysis was performed by means of unpaired '*t*' test applying the Bonferroni's correction. Stepwise multiple linear regression was performed to evaluate the relative importance of age, disease duration, GH and IGF-I levels, values of systolic and diastolic blood pressure, presence or absence of glucose tolerance abnormalities, fibrinogen levels and lipid profile on structural (IMT) and functional (RI) parameters. The χ^2-test was also used where appropriate.

Results

Clinical study (Table 1)

BMI was similar in active and cured patients with acromegaly and in controls (Table 1): none of the 75 subjects was obese while 10 patients with active disease (33·3%), six patients cured from acromegaly (40%) and six controls (20%) were overweight ($\chi^2 = 2{\cdot}31$, $P = 0{\cdot}3$). SBP values, but not DBP, were significantly higher in patients with active disease than in cured patients and controls ($P = 0{\cdot}003$). Hypertension was

Table 2 Results of echodoppler ultrasonography in acromegalic patients and controls

Parameters	Acromegalic patients		Controls	*P*
	Active disease ($n = 30$)	Cured ($n = 15$)	($n = 30$)	
At the common carotid artery level				
Intima-media thickness (mm)				
Right	1·04 ± 0·04	1·04 ± 0·05	0·8 ± 0·02*	0·000
Left	1·1 ± 0·04	1·1 ± 0·06	0·8 ± 0·01*	0·000
Systolic peak velocity (cm/s)				
Right	90·0 ± 5·6†	78·0 ± 6·0	64·5 ± 4·5	0·002
Left	87·0 ± 4·8†	82·0 ± 6·7	65·4 ± 5·4	0·009
Diastolic peak velocity (cm/s)				
Right	20·5 ± 1·4	19·2 ± 0·9	19·0 ± 1·4	0·6
Left	23·4 ± 2·4	20·9 ± 1·4	19·8 ± 1·7	0·5
Resistance index				
Right	0·8 ± 0·13	0·8 ± 0·14	0·6 ± 1·9	0·9
Left	0·8 ± 0·09	0·8 ± 0·07	0·7 ± 1·0	0·9
At the internal carotid artery level				
Systolic peak velocity (cm/s)				
Right	67·6 ± 3·6	67·6 ± 4·6	64·5 ± 4·5	0·8
Left	67·9 ± 3·7	60·4 ± 4·4	65·4 ± 5·4	0·6
Diastolic peak velocity (cm/s)				
Right	22·1 ± 1·5	20·6 ± 1·8	19·0 ± 1·4	0·3
Left	22·1 ± 1·6	20·1 ± 1·4	19·8 ± 1·7	0·5
Resistance index				
Right	0·7 ± 0·02	0·7 ± 0·02	0·7 ± 0·01	1
Left	0·6 ± 0·01	0·7 ± 0·03	0·7 ± 0·06	0·8
Carotid Ratio				
Right	1·18 ± 0·09	1·06 ± 0·08	0·99 ± 0·09	0·3
Left	1·04 ± 0·05	0·93 ± 0·09	1·01 ± 0·08	0·6

*Significant *vs.* active and cured patients; †significant *vs.* cured patients and healthy controls.

found in nine (30%) patients with active disease, in two (13·3%) of those cured from acromegaly and in none of controls ($\chi^2 = 10{\cdot}81$, $P < 0{\cdot}004$). Among these 11 patients, hypertension was mild in seven and moderate in four patients.

Metabolic study (Table 1)

Fasting blood glucose levels were significantly higher both in patients with active disease and in those cured from the disease than in controls ($P < 0{\cdot}001$). Circulating insulin levels were significantly higher in patients with active disease than in cured patients and controls ($P < 0{\cdot}001$). A significant increase in insulin response to glucose load was found in patients with acromegaly as compared to controls, either when evaluated as peak (206·4 ± 43·8 *vs.* 102·1 ± 19·1 *vs.* 42·8 ± 10·1 μU/l) or as AUC (Fig. 1). No difference was found in the glucose peak after glucose load (183·3 ± 13·5 *vs.* 177·0 ± 12·4 *vs.* 159·1 ± 11·2 mg/dl) while glucose AUC was significantly higher in acromegalic patients than in controls (Fig. 1). Diabetes mellitus was found in five (16·6%) patients with active disease, in one (6·6%) patient with inactive disease and in none of the controls ($\chi^2 = 5{\cdot}7$, $P = 0{\cdot}058$). Reduced glucose tolerance was found in eight (26·7%) patients with active disease, three (20%) patients with inactive disease and in four (13·3%) controls ($\chi^2 = 1{\cdot}7$, $P = 0{\cdot}4$). As a whole, glucose tolerance abnormalities were found in 13 (43·3%) patients with active disease, in four (26·6%) patients with inactive disease and in four controls (13·3%) ($\chi^2 = 6{\cdot}71$, $P = 0{\cdot}03$). Total blood cholesterol levels were similar in the three groups whereas LDL-cholesterol and triglycerides levels were significantly higher in patients cured from the disease than in controls (Table 1). HDL-cholesterol levels were significantly lower both in patients with active disease and in those cured from the disease than in controls ($P < 0{\cdot}001$). Hypercholesterolaemia was found in

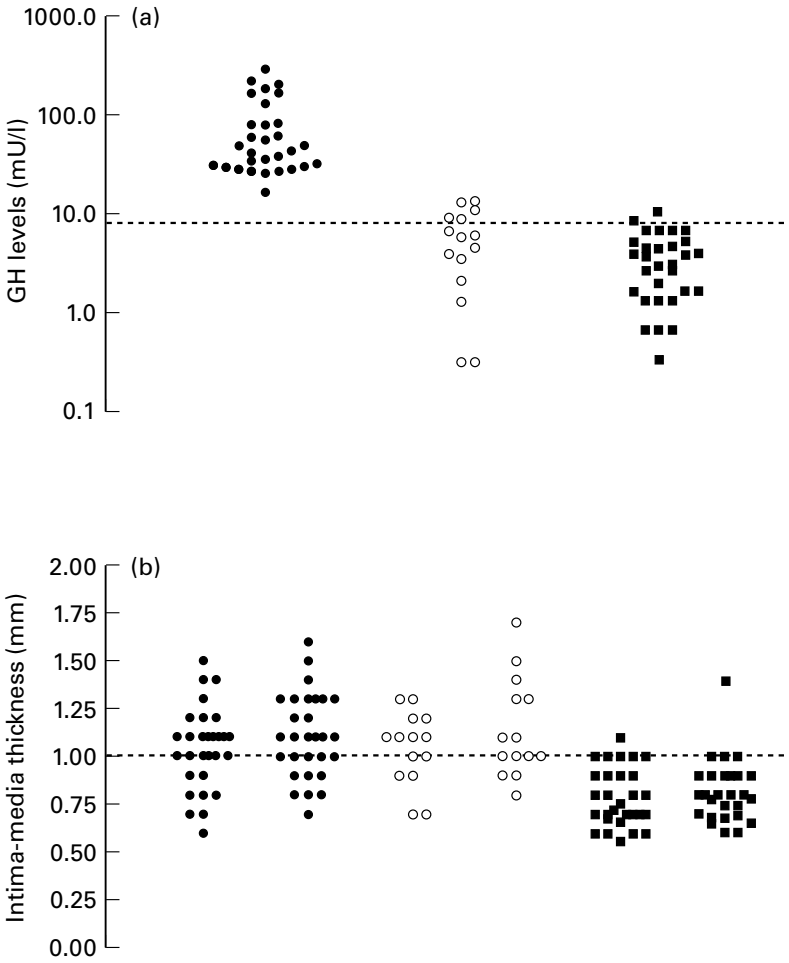

Fig. 2 Individual data of GH levels (a) and intima-media thickness (b) measured at the level of right and left common carotid arteries in patients with active (●) and cured (○) acromegaly and controls (■). The shaded lines indicate the normal GH and IMT value.

four (13·3%) patients with active disease, one (6·7%) patient with inactive disease and in one (3·3%) control ($\chi^2 = 2{\cdot}1$, $P = 0{\cdot}3$). Hypertriglyceridaemia was found in one (3·3%) patient with active disease, two (13·3%) patients with inactive disease and in none of controls ($\chi^2 = 4{\cdot}7$, $P = 0{\cdot}09$). Serum fibrinogen levels were significantly higher both in patients with active disease and in those cured from the disease than in controls ($P < 0{\cdot}001$). Hyperfibrinogenaemia was found in 18 (60%) patients with active disease, nine (60%) patients with inactive disease and in none of controls ($\chi^2 = 28{\cdot}1$, $P < 0{\cdot}001$). No difference was found in PT and APTT levels among the three groups.

Vascular study (Table 2)

At the level of right and left common carotid arteries, IMT was significantly higher both in patients with active disease and in those cured from the disease than in controls (Fig. 2; $P < 0{\cdot}001$). A significant increase of IMT (> 1 mm) at one or both common carotid arteries was found in 21 patients with active disease (70%) ranging between 1·1 and 1·6 mm, in nine patients cured from the disease (60%) ranging between 1·1 and 1·7 mm and in two controls (6·6%) ranging 1·1–1·4 mm ($\chi^2 = 28{\cdot}9$, $P < 0{\cdot}001$). Both right and left SPV, but not DPV, were significantly higher in patients with active disease than in those cured from the disease and in controls ($P < 0{\cdot}01$). No difference was found in the RI among the three groups (Table 2). Well defined carotid wall plaques were detected in two patients (6·6%) with active disease, in one patient cured from the disease (6·6%) and in two controls (6·6%). All these subjects were above 60 years of age (Table 3) ($\chi^2 = 0$, $P = 1$).

At the level of right and left internal carotid arteries, SPV, DPV and RI were similar among the three groups (Table 2). Well defined carotid wall plaques were detected in three patients with active disease (10%), two patients cured from the disease (13·3%) and in one control (3·3%) (Table 3) ($\chi^2 = 1{\cdot}63$, $P = 0{\cdot}4$).

The ratio between DPV of right and left internal *vs.* common

Table 3 Antropometric, hormonal and ultrasonographic findings of the six subjects showing well defined plaques at the level of both common or internal carotid arteries

Patient (sex/age)	Mean GH level (mU/l)	IGF-I level (μmol/l)	Disease duration (years)	DBP (mmHg)	Glucose level (mg/dl)	Insulin level (pmol/l)	Triglyceride level (mg/dl)	Total cholesterol level (mg/dl)	Fibrinogen level (μmol/l)M	US-findings of atherosclerotic plaques	
										Site	Size (length x thickness, mm)
Patients with active acromegaly											
M/39	44	72·0	15	110	106	358·8	234	298	14·6	R and L IC	2·1 × 0·6, 2·3 × 0·5
F/40	212·5	75·5	5	80	125	179·4	216	173	11·0	R and L IC	0·6 × 0·5, 0·5 × 0·7
M/43	175	198·7	8	90	118	222·4	71	250	13·6	L IC	0·5 × 0·6
F/63	47·5	50·8	20	100	109	179·5	90	210	15·6	R CC	0·7 × 0·4
F/65	37·5	63·1	10	110	111	265·5	107	250	14·4	L CC	0·3 × 1·5
Patients cured from acromegaly											
F/30	7·0	24·2	15*	90	75	107·6	75	228	10·0	L IC	0·3 × 0·4
M/60	5·5	35·7	15*	95	92	37·3	50	232	15·8	R and L CC, L CI	2·1 × 0·5, 0·6 × 0·2, 1·7 × 0·5
Controls											
F/60	0·5	20·0	–	95	98	78·9	80	198	9·2	R CC	1·0 × 1·5
M/63	2·0	22·9	–	90	110	93·3	100	175	8·5	R and L CC, R CI	1·5 × 0·5 0·9 × 0·6, 1·4 × 0·9

DBP, diastolic blood pressure, US, ultrasonographic, R, right, L, left, CC, common carotid, IC internal carotid.
Fasting glucose and insulin levels are shown. *For cured patients the disease duration refers to both the active and inactive disease period: for the female patient 10 years of active and 5 years of inactive disease and for the male 5 years of active and 10 years of inactive disease.

carotid arteries was similar among the three groups. In detail, seven patients with active disease (23·3%), one cured from acromegaly (6·6%) and six controls (20%) had ratio values above 1·5: five active patients, the cured one and four controls had a mild stenosis, two active patients and two controls had a moderate stenosis.

Correlation analysis

At linear correlation analysis, a strong correlation between right and left common carotid arteries IMT was found ($r = 0{\cdot}789$, $P < 0{\cdot}001$). IMT was significantly correlated to age ($r = 0{\cdot}33$; $P < 0{\cdot}01$), GH ($r = 0{\cdot}30$, $P < 0{\cdot}01$), IGF-I ($r = 0{\cdot}34$, $P < 0{\cdot}05$), fibrinogen ($r = 0{\cdot}5$, $P < 0{\cdot}001$), glucose ($r = 0{\cdot}3$, $P < 0{\cdot}05$), insulin ($r = 0{\cdot}39$, $P < 0{\cdot}001$), triglycerides ($r = 0{\cdot}29$, $P < 0{\cdot}01$), and HDL-cholesterol ($r = -0{\cdot}44$, $P < 0{\cdot}001$) levels. The results of the multistep correlation analysis are shown in Table 4. The strongest predictor of common carotid IMT was fibrinogen level, followed by age and GH level, while the strongest predictors of common and internal carotid RI were IGF-I and GH levels, respectively.

Discussion

The results of the current study demonstrated that a significant increase in IMT of right and left common carotid arteries was found both in patients with active acromegaly and in those cured from acromegaly as compared to age-and sex-matched controls. However, no increase in the prevalence of well defined atherosclerotic plaques was found either in patients with active or cured acromegaly as compared to controls despite the expected evidence of an increased prevalence of glucose tolerance abnormalities, hypertension, hypercholesterolaemia and hyperfibrinogenaemia in patients with active

Table 4 Results of multiple regression analysis

Dependent variables	Independent variables	R	R^2	*P*
Common carotid IMT				
	Fibrinogen levels	0·511	0·261	< 0·001
	Age	0·596	0·355	< 0·001
	GH levels	0·631	0·398	0·033
Common carotid RI				
	IGF-I levels	0·279	0·078	0·02
Internal carotid RI				
	GH levels	0·388	0·151	0·047

IMT, intima-media thickness (mm), RI, resistance index, ratio (see methods).

acromegaly. Moreover, both GH and IGF-I levels were among the strongest predictors of structural and functional parameters of both common and internal carotids.

In recent years, a large body of evidence has been accumulated on the detrimental effect of chronic GH and IGF-I excess on cardiac function in acromegalic patients (Saccà *et al*., 1994; Colao & Lombardi, 1998; Isgaard *et al*., 1998). The concomitant presence of hypertension and glucose tolerance abnormalities further worsened cardiac function in acromegaly (Lopez-Velasco *et al*., 1997; Colao *et al*. 2000). In addition, a heterogeneous distribution of cardiac output has been recently demonstrated in patients with active acromegaly by a direct measurement of brachial artery haemodynamics which showed lower regional blood flow and increased local resistance (Chanson *et al*., 1998).

However, despite the evidence of morphological alterations in the peripheral microcirculation (Schiavon *et al*., 1999), the vascular component of these severe complications of chronic GH and IGF-I excess has been never investigated. In contrast, this issue had been carefully addressed in patients affected with GH deficiency (GHD) who were demonstrated to have a premature mortality due to cardiovascular disease (Rosén & Bergtsson, 1990; Bülow *et al*., 1997). A significant increase in the prevalence of premature atherosclerosis was reported in adult patients with GHD (Markussis *et al*., 1992; Borson-Chazot *et al*., 1999; Pfeifer *et al*., 1999). A significant increase in IMT at the level of both common carotid arteries was reported either in patients with GHD developed during childhood or adulthood (Markussis *et al*., 1992; Capaldo *et al*., 1997; Borson-Chazot *et al*., 1999; Pfeifer *et al*., 1999). In addition, a beneficial effect of GH replacement on vascular abnormalities has been recently demonstrated as GH treatment in hypopituitary GHD men reversed early morphological and functional atherosclerotic changes in major arteries (Pfeifer *et al*., 1999). The reduction in IMT observed in GH treated GHD patients occurred without a consensual improvement of cardiovascular risk factor parameters, as only a transient 10% decrease in LDL-cholesterol levels was found after 6 months (Borson-Chazot *et al*., 1999). These findings suggested a parietal effect of GH on the arterial wall (Bots *et al*., 1997). As further support to this hypothesis, GH levels strongly predicted IMT in the 75 subjects enrolled in the present study. However, most of the physiological effects of GH on the vascular system are mediated by IGF-I.

IGF-I has long been associated with growth and differentiation of whole body organs and tissues. Beyond these effects and those metabolic insulin-like, such as glucose uptake, glycogen synthesis and amino-acid transport, IGF-I may contribute to the regulation of vascular tone (Guler *et al*., 1989; Hirshberg *et al*., 1993; Copeland & Sreekuran, 1994). Endothelial cells possess high-affinity binding sites for IGF-I (Delafontaine *et al*., 1991) and IGF-I has been shown to increase endothelial NO formation (Tsukahara *et al*., 1994; Böger *et al*., 1996). Decreased NO activity is associated with impaired arterial vasodilator capacity, increased platelet aggregability and intimal thickening (Böger *et al*., 1996). An inverse relationship between free IGF-I levels and IMT has been recently reported in healthy elderly subjects (Janssen *et al*., 1998).

The results of the present study showed a significant increase of IMT, without any increase in the prevalence of well-defined atherosclerotic plaques, in active acromegalic patients. In addition, a similar finding was also observed in patients cured from acromegaly, indicating that even a long period of GH/IGF-I suppression was not able to reverse intimal thickening. On the other hand, the prevalence of other abnormalities able to increase the cardiovascular risk, such as glucose tolerance alterations, hypertension, unfavourable lipid profile and increased fibrinogen levels, was still slightly higher in cured acromegalic patients than in controls. An adaptive increase of IMT, unrelated to atherosclerosis, has been suggested as a compensatory response of the vessels to changes in blood flow or vascular tone (Bots *et al*., 1997). In fact, in our 75 subjects age was one of the strongest predictors of IMT as well as GH levels. The apparent contrast between the increase in IMT observed in acromegalic patients and the reported decrease in GHD patients following GH replacement could be due to the pathological overgrowing effect of chronically elevated GH levels in acromegaly. The consensual increase of circulating IGF-I together with the locally produced IGF-I (Delafontaine *et al*., 1991; Tsukahara *et al*., 1994) can, however, protect from the formation of atherosclerotic plaques despite the presence of low HDL-cholesterol and high fibrinogen levels. Furthermore, acromegalic patients have an increase in insulin levels which are known to be directly correlated with IMT (Kahn *et al*., 1993; Sowers *et al*., 1993; Wu *et al*., 1994). However, unlike insulin, IGF-I may potentially be a more important regulator of regional blood flow: IGF-I was demonstrated to possess a direct impact on the vasculature and attenuates contractility in rat arteries, an effect related to NO production (Gryglewski *et al*., 1986). Furthermore, infusions of recombinant human IGF-I into both humans and rats increase renal blood flow and decrease vascular resistance in that organ (Guler *et al*., 1989; Hirshberg *et al*., 1993). As IGF-I is not only secreted by vascular smooth muscle cells but also stimulated vascular NO production, it was suggested that this hormone play a significant paracrine/autocrine role in the regulation of local blood flow (Delafontaine *et al*., 1991).

In conclusion, besides the increased prevalence of hypertension, glucose tolerance abnormalities, insulin resistance,

unfavourable lipid profile and high fibrinogen levels, an increase of IMT, which seems to indicate an early atherosclerosis and is considered as a risk factor for myocardial infarction and stroke in adults (O'Leary *et al*., 1999), but without any increase in the prevalence of well-defined atherosclerotic plaques was found in acromegalic patients. Long-term achievement of safe GH and IGF-I levels indicating cure from the disease did not significantly modify cardiovascular risk factors and IMT in acromegaly. Although prospective studies are needed before definitive conclusions can be drawn, these data suggest that the increased mortality reported by epidemiological studies in acromegaly is more likely to be due to a direct heart alteration than to a vascular damage.

Acknowledgements

This study was partially supported by grant n.9906153187 from MURST Rome, Italy.

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