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Evidence of Cognitive and Neurophysiological Impairment in Patients with Untreated Naive Acromegaly

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Context: Recent studies have suggested that long-term exposure to high levels of GH and IGF-I affect brain and cognitive functions. However, very few human studies have challenged this hypothesis.

Objective: The aim of this study is to explore whether GH/IGF-I excess in naive patients with acromegaly alters cognitive functions, particularly memory, and whether these alterations are accompanied by neurophysiological correlates.

Design: We conducted a comprehensive neuropsychological and neurophysiological exam on 16 naive acromegaly patients and 16 strictly matched healthy controls. Comparative analyses were carried out on major neurocognitive domains (executive functions, visual/verbal memory, attention, visuoconstructive abilities, and verbal fluency) and on quantitative electroencephalogram and low-resolution brain electromagnetic tomography sources. Results were correlated with GH and IGF-I hormone concentrations.

Results: Short- and long-term memory were the most severely impaired cognitive functions. Moreover, memory performance correlated negatively with GH and IGF-I concentrations. No association was detected between depression and memory impairment, and only a marginal association was found with quality of life. Finally, acromegaly patients showed power attenuation in fast frequency electroencephalogram bands, as well as decreased activity in prefrontal and middle temporal cortices, that was associated to cognitive performance.

Conclusions: Results provide evidence of cognitive and neurophysiological impairment, characterized by moderate-to-severe memory impairment and decreased neural activity in specific brain areas. High levels of GH and IGF-I in acromegaly patients could be the basis for these findings. (***J Clin Endocrinol Metab*** **95: 0000–0000, 2010)**

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Abbreviations: AcroQoL, Acromegaly Quality of Life Questionnaire; BDI-II, Beck Depression Inventory-II; CI, contamination index; CV, coefficient of variation; DLPFC, dorsolateral prefrontal cortex; EEG, electroencephalogram; LMW-R, Luria's Memory Word Test-Revised; LORETA, low-resolution brain electromagnetic tomography; QEEG, quantitative EEG; QoL, quality of life.

Acromegaly is a rare disease that results when overfunctioning somatotropic pituitary cells produce excess GH. This excess of IGF-I is found in the blood and GH-target tissues. The disease typically follows an insidious clinical course spanning several years before being diagnosed, usually in middle age. Given this development, clinical signs and symptoms of acromegaly often appear after prolonged oversaturation of GH and IGF-I (1). Although the effects of long-term exposure to high levels of GH and IGF-I on morbidity and mortality are well known, they have yet to be established in regard to the human brain. Patients with acromegaly could provide a clinical model for studying the noxious effects of supraphysiological GH/IGF-I levels on the brain and cognition.

The GH and IGF-I axis is involved in normal brain functioning, actively participating in the brain's neurogenesis, myelinization, and biochemistry (2). Certain brain areas contain a high number of GH/IGF-I binding sites, namely choroid plexus and hypothalamus, but the human hippocampus stands out, where GH receptor expression is 2- to 4-fold higher (3). This area is essential to higher cognitive functions, such as learning and episodic memory. IGF receptors are also abundant in neocortical and striatum areas (4).

The finding of GH and IGF-I in cerebrospinal fluid and its brain-binding sites—namely the hippocampus and neocortical areas—suggests that the GH/IGF-I axis affects cognition (5), although little is known regarding its neurophysiological and cognitive effects (6). Alteration of this axis, particularly in patients with GH deficiency, alters numerous cognitive functions, namely memory and executive functions (7). Recent research on acromegaly, using a cognitive electroencephalogram (EEG)-evoked potential paradigm, found a significant P300 amplitude reduction compared with patients with GH deficiency and healthy controls (8). Magnetic resonance imaging studies have also shown an abnormal increase in gray and white brain matter volumes at the expense of cerebrospinal fluid (9). Interestingly, these authors found a higher prevalence of patients with white matter lesions that could be related to the metabolic and vascular comorbidities of acromegaly.

The aim of this study was to determine whether patients with naive acromegaly and long-term exposure to supraphysiological levels of GH and IGF-I had alterations in their central nervous system. We conducted a comprehensive neuropsychological and neurophysiological assessment of major neurocognitive domains, using neuropsychological tasks proven useful in detecting cognitive impairment due to organic causes. Neurophysiological correlates for the neuropsychological assessment results were explored. Special attention was given to the physiological link between GH/IGF-I and memory functions. Mood and quality of life (QoL), were also assessed, given that cognitive functions in this population may be affected by psychopathological and QoL abnormalities (10, 11).

Subjects and Methods

Subjects and procedures

Seventeen patients with newly diagnosed acromegaly were enrolled in the current study. Patients were recruited from March 2008 through July 2009 from four university hospitals located in Andalusia, Spain. The acromegaly suspicion was based on the presence of relevant clinical features, and the diagnosis was confirmed by failure of plasma GH to reduce to levels less than 1 μg/ml during a 75-g oral glucose tolerance test. Median (percentiles 75th–25th) GH and IGF-I were 9.3 ng/ml (22.07–5.32) and 747.5 ng/ml (1226.7–640). IGF-I level was greater than 3 SD values above the age-specific normal range [median (percentiles) = 10 (13.5–7.65)] (Table 1). Once the diagnosis was confirmed, routine pituitary function assessments were done to evaluate the presence of hypopituitarism [serum T_4, TSH, 0900-h cortisol, prolactin (PRL), LH, FSH, testosterone, or estradiol]. Table 1 shows patients' clinical characteristics.

Patients were prescreened for major psychiatric disorders (schizophrenia, schizoaffective disorder, bipolar and unipolar affective disorder) or neurological disease background. No patient was excluded due to major psychiatric disorder. One patient, aged 72 yr, was discarded from the study sample for scoring less than 24 in the Mini-Mental State Examination, suggesting mild cognitive impairment. Sixteen patients [12 females, 4 males; median age, 36 yr (range, 24–59)] gave written consent to participate in the study after reading a consent form. Cases received standard substitution therapy for thyroid and adrenal hypofunction as needed. No patient had received somatostatin analog or dopamine agonist treatment before the study. Height, weight, body mass index, blood count, standard biochemical analysis, and hormonal parameters were obtained in the fasting state on the day of study participation (between 0800 and 0900 h). PRL level was again assessed at the study time point, given that hyperprolactinemia was present in three patients, and it is documented that it may cause attention and memory impairment (12).

Serum GH concentrations were measured using a time-resolved fluoroimmunometric assay (Wallac, Turku, Finland). Intraassay coefficients of variation (CVs) for GH levels ranging from 2.6 to 17 ng/ml were 5.0 and 5.6%; interassay CVs for these levels were 5.0 and 8.5%. Serum IGF-I levels were measured by a commercial immunoradiometric assay kit after acid ethanol extraction (Immunotech SA, Marseilles, France), with an interassay CV of 3.5% (at an IGF-I level of 77 ng/ml) to 6.8% (at an IGF-I level of 1358 ng/ml). The intraassay CV were 5.0 and 8.0%, respectively. Serum PRL was measured by a fluoroimmunoassay (Autodelfia hPRL kit; Wallac, Inc., Turku, Finland), with an interassay CV of 3.5 and 3.7% (at PRL levels from 14.1 to 33.7 ng/ml). The intraassay CVs were 3.5 and 2.7%, respectively. Serum cortisol was measured by a fluoroimmunoassay (Autodelfia hLH kit, Wallac, Inc.). The interassay and intraassay CVs at a cortisol level from 129 to 899 nmol/liter were 2.8 and 2.0% and 3.0 and 1.9%, respectively. Serum free testosterone, estradiol, LH, FSH, ACTH, T_4, and TSH measurements are described elsewhere (13).

AQ:E
AQ:F
AQ:G
T1, AQ:H-I
AQ:J

TABLE 1. Demographic and clinical data on acromegaly patient sample

Patient no.	Gender	Age (yr)	Years of education	GH levels (ng/ml)	IGF-I levels (ng/ml)	IGF-I (SD)	PRL (ng/ml)	Comorbidities/treatment, dosage	Tumor size
1	M	51	>16	17.90	795	11.9	19.07	Myocardiopathy (myocardial infarction, coronary artery stints)/aspirin, 150 mg/d. Hyperlipidemia/simvastatin, 20 mg/d. Hypertension/carvedilol, 37.5 mg/d, and losartan, 50 m/d.	Microadenoma (8 mm)
2	F	29	12	10.50	1925	26.7	50.99	No/no	Microadenoma (5 mm)
3	F	36	14	14.00	804	10.9	61.08	No/no	Macroadenoma (20 mm) with invasion into cavernous and sphenoidal sinus
4	M	25	>16	36.9	1315	17.0	123.73	No/no	Noninvasive macroadenoma (30 mm)
5	F	30	8	6.34	663	7.8	69.93	No/no	Noninvasive macroadenoma (25 mm)
6	F	37	10	4.20	612	7.6	4.99	Hypertension/losartan, 50 mg/d PO	Noninvasive macroadenoma (15 mm)
7	F	59	7	7.30	645	9.6	11.90	Hypertension/●●●, 5 mg bid PO. Hypopituitarism/hydrocortisone, 20 mg bid PO; and L-T_4, 75 μg/d PO. OSAS/CPAP. Hyperlipidemia/pravastatin, 20 mg/d PO.	Noninvasive macroadenoma (15 mm)
8	F	24	7	6.83	639	5.6	18.88	No/no	Invasive macroadenoma (18 mm) to sphenoidal sinus
9	F	36	8	5.00	567	6.8	n.a.	No	Noninvasive macroadenoma (18 mm)
10	F	57	7	30.10	1438	24.8	5.33	Hypertension/enalapril, 5 mg bid PO	Noninvasive macroadenoma (28 mm)
11	M	37	>16	22.60	962	13.6	24.40	Sleep apnea/CPAP	Noninvasive macroadenoma (15 mm)
12	M	32	8	20.50	790	9.9	16.38	Hypopituitarism/hydrocortisone, 20 mg bid PO. Bilateral hemianopsia/L-T_4, 75 μg/d PO.	Macroadenoma (30 mm) with suprasellar extension
13	F	35	7	10.60	818	11.1	21.78	Carpal tunnel syndrome and euthyroid diffuse goiter	Macroadenoma (20 mm) with suprasellar extension
14	F	56	14	4.14	414	5.2	5.47	Diabetes mellitus type 2/metformin, 850 mg three times daily; and Lantus, 30 U/d sc. Hypertension/enalapril, 5 mg bid PO. Sleep apnea/CPAP. Hypopituitarism/L-T_4, 100 μg/d PO. Hyperlipidemia/simvastatin, 20 mg/d PO.	Noninvasive macroadenoma (21 mm)

(*Continued*)

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TABLE 1. Continued

Patient no.	Gender	Age (yr)	Years of education	GH levels (ng/ml)	IGF-I levels (ng/ml)	IGF-I (SD)	PRL (ng/ml)	Comorbidities/treatment, dosage	Tumor size
15	F	45	7	17.90	871	13.2	28.12	Diabetes mellitus type 2/NovoMix, 30/70 U/d sc and Lantus 30 U/d. Hypertension/losartan, 50 mg bid PO. Sleep apnea/CPAP. Hypopituitarism/L-T_4, 100 μg/d PO. Hyperlipidemia/atorvastatin, 80 mg/d PO. Carpal tunnel syndrome. Bilateral hemianopsia.	Macroadenoma (43 mm) with suprasellar extension
16	F	27	8	7.89	726	7.9	82.11	No/no	Macroadenoma (25 mm) with suprasellar extension

Blood samples were collected on the same day as neurophysiological and neuropsychological assessments. M, Male; F, female; OSAS, obstructive sleep apnea syndrome; CPAP, continuous positive airwave pressure; bid, twice daily; PO, orally; N/A, not applicable.

The control group included 16 demographically matched right-handed healthy subjects. Inclusion criteria for this group were the absence of current systemic disease, or a history of endocrinological disease, or present or previous drug abuse. These subjects were prescreened for neurological impairment and major psychiatric disorders. Analytical evaluations were not performed on healthy controls. Gender and years of formal education were strictly matched. A difference of ±2 yr was accepted for age matching. No significant differences were found in the age variable ($P = 0.98$; patients' mean age = 38.50 yr; controls' mean age = 38.92 yr).

Neurocognitive, emotional, and QoL assessments

Both groups underwent a comprehensive neurocognitive assessment of major cognitive domains. The tests and tasks included in this assessment have proven sensitive to cognitive impairment due to organic causes. A classification was used to subdivide the tests into major cognitive domains: attention, executive functioning, short- and long-term memory (verbal and visual), visuoconstructive ability, and language skills (verbal fluency) (14).

Basic attention processes, namely vigilance and sustained attention, were assessed using a computerized version of the letter cancellation task, included in the Seville Neuropsychological Battery (15). Psychomotor speed and visual scanning abilities were measured using the Trail Making Test (part A). Executive functions were assessed with a battery of neuropsychological tests. The Trail Making Test (part B) was used to measure cognitive flexibility and visuoconceptual tracking. The Stroop Color-Word Test was used to assess inhibitory function. The Tower of Hanoi-Sevilla, a complex problem-solving task, assesses planning, goal-directed behavior, and error learning.

Memory was assessed using a battery of tests that measured short- and long-term memory (verbal and visual) and learning processes. The verbal short-term memory and learning processes were assessed using the immediate condition of the Luria's Memory Words Test-Revised (LMW-R). Ten indexes are calculated for this task, and they were used to score the immediate condition (description and calculation of memory indexes provided by LMW-R can be found in Ref. 16). Working memory was assessed using the forward and backward versions of the Digit Span Test, included in the Wechsler Memory Scale-Third Edition (17). Verbal and visual delayed recall was assessed using the delayed recall of the LMW-R and the memory condition of the Complex Figure Test, respectively.

Visuocontructive abilities were assessed using the copy condition of the Complex Figure Test. The Controlled Oral Word Association Test was used to measure verbal fluency (18).

The neurocognitive assessment was conducted between 0900 and 1100 h, after the neurophysiological exam. The assessment usually lasted 90 min, with a 10-min break halfway through. Tasks and tests were administered randomly across subjects, excluding the delayed recall condition of the LMW-R. This condition was inserted three tasks after a learning condition (approximately 30 min). Neuropsychological tests were administered by trained neuropsychologists from the University of Seville Human Neuropsychology Laboratory. The same examiners evaluated both patients and controls.

We used two questionnaires to assess the impact of acromegaly on mood and QoL. The Beck Depression Inventory-II (BDI-II) (19) is a widely used tool that assesses the emotional state of the neuropsychiatric population (20). We used BDI-II cutoffs to

classify data into no depression, mild depression, moderate depression, and severe depression (21). The Acromegaly Quality of Life Questionnaire (AcroQoL), specifically developed for QoL assessment in patients with acromegaly (22), was used in our study. This questionnaire consists of 22 statements, some assessing frequency of occurrence (from always to never) and others assessing level of agreement (from total agreement to total disagreement). The statements in this questionnaire evaluate both the physical and psychological aspects of acromegaly.

Quantitative EEG (QEEG) assessment

AQ: K

EEG was recorded using a clinical EEG unit (Truscan 32; Deymed Diagnostic, Payette, ID). We obtained EEG data from a 3-min recording on patients and controls at awakening from resting state (linked ears reference; bandwidths between 0.1 and 100 Hz; sampling frequency = 256 Hz). Nineteen electrodes were placed according to the international 10–20 system: seven frontal locations (Fp1, Fp2, F7, F3, Fz, F4, F8), three central (C3, Cz, C4), four temporal (T3, T4, T5, T6), three parietal (P3, Pz, P4), and two occipital (O1, O2). Participants were asked to close their eyes and remain relaxed and alert while EEG activity was recorded. All recordings were performed between 0800 and 1000 h.

EEG data were analyzed and fragmented into consecutive 2-sec epochs. Only artifact-free epochs were considered for further analyses. Within each recording, tests for split-half and test retest reliability were conducted on the edited EEG segments. Only records with more than 90% reliability entered into the spectral analyses. Data preprocessing and filtering were carried out offline in MATLAB (version 7.0) using EEGLAB (http://www.sccn.ucsd.edu/eeglab/index.html) (23) and custom scripts. Power spectra (1–40 Hz, resolution of 1 Hz) were calculated using fast Fourier transform and were averaged across frequency bands and scalp locations, namely frontal (FP1, FP2, F3, Fz, F4, F7, F8), central-temporal (C3, Cz, C4, T3, T4, T5, T6), and parietal-occipital (P3, Pz, P4, O1, O2).

EEG source localization

Low-resolution brain electromagnetic tomography (LORETA) was used for thes localization of cortical sources of EEG activity (24). This solution procedure has been extensively validated (25, 26). LORETA can be used on data collected by low spatial sampling of the 10–20 System (19 electrodes), when cortical sources are estimated from resting EEG rhythms (25). The LORETA version employed in this study uses a three-shell spherical head model that includes scalp, skull, and brain compartments. LORETA procedure estimates the distribution of absolute current density (numerically and visually using scaled color intensity) for brain electrical activity and displays on a dense grid of 2394 voxels based on the digitized Talairach Magnetic Resonance Imaging Atlas (27). Calculation of LORETA is limited to cortical gray matter and hippocampus and produces a spatial resolution of approximately 1–2 cm.

Ethics

The study was approved by the Ethics Committee of the Virgen del Rocío Hospital. The purpose of the study was carefully explained to patients and relatives and healthy controls, who were provided with written information on the background of the study. Participants who agreed then signed a written consent form.

Data analysis

Median and percentiles for demographic, clinical, and neurocognitive variables were obtained for each group. Mann-Whitney two-sample tests were applied to compare each group's performance in the neurocognitive, depression, and QoL measures. Spearman's nonparametric correlations were applied to study possible relationships between depression, QoL, and hormonal and neuropsychological data. Correlations between IGF-I and neuropsychological variables were conducted using both absolute and IGF-I SD scores (28). The Holm-Bonferroni method was used to adjust the overall nominal significance level for multiple comparisons and correlations.

A 2 × 2 ANOVA for repeated measures was conducted to evaluate performance differences between patients and healthy subjects in the LMW-R learning condition (measured by the number of correct words in the last trial of this condition) and delayed recall condition. We chose the number of correct words recalled after the last trial of the learning condition as representative of this condition because it normally represents the subject's best performance. Bonferroni corrections were applied to protect against type I error when performing multiple comparisons.

Effect sizes, weighted for SD, were also obtained for each comparison (Cohen's *d*) (29), showing the size of any observed differences. The 95% confidence intervals were calculated for each comparison. Statistical analyses were performed using SPSS 13.0 software (SPSS Inc., Chicago, IL).

For QEEG data, Mann-Whitney tests for independent measures were used to compare the differences between EEG variables in the two clinical conditions. Level of significance when performing simultaneous multiple comparisons was corrected employing rough false discovery rate.

The frequency bands of interest for the LORETA analyses were δ (1–3 Hz), θ (4–7 Hz), α (8–12 Hz), $\beta 1$ (13–18 Hz), $\beta 2$ (19–21 Hz), $\beta 3$ (22–30 Hz), γ (31–40 Hz), and full band (1–40 Hz). A subject-wise normalization was applied. LORETA images of the two study groups were statistically compared using voxel-by-voxel *t* test. We also calculated exact randomization probabilities (5000 randomizations), corrected for multiple comparisons. The threshold for determining statistical significance was set at $P < 0.05$.

Results

Neuropsychological results

T2, AQ:L-M

Table 2 summarizes neuropsychological results for both groups. Patients' performance did not differ significantly from healthy controls in tasks assessing attention and executive functions. Memory assessment showed significant differences for both verbal and nonverbal content. Patients showed a higher contamination index (CI) in LWM-R ($P < 0.005$) and a smaller short-term memory span in the backward digit span test ($P = 0.007$), and they recalled fewer details in the Complex Figure Test memory condition ($P < 0.0005$). According to Cohen's interpretation of effect size (29), these differences are large (Cohen's *d* for CI comparison = 2.57; forward digit span comparison = 2.65; Complex Figure Test memory con-

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TABLE 2. Neuropsychological performance of acromegaly patients and healthy controls

	Acromegaly patients (n = 16)	Matched controls (n = 16)	*P* value
Attention			
Simple attention test (% correct)	98.93 (100, 98.00)	100.00 (100, 100)	0.496
Simple attention test (RT in sec)	0.45 (0.52, 0.42)	0.46 (0.50, 0.39)	0.724
Conditional attention test (% correct)	98.01 (100, 95.00)	100.00 (100, 96.00)	0.336
Conditional attention test (RT in sec)	0.38 (0.44, 0.35)	0.38 (0.45, 0.35)	0.960
Trail Making Test A (time in sec)	40.00 (52.75, 31.00)	37.00 (44.50, 25.05)	0.101
Trail Making Test A (errors)	0	0	1
Executive functioning			
Trail Making Test B (time in sec)	105.50 (235.70, 78.50)	75.00 (107.50, 60.50)	0.092
Trail Making Test B (errors)	0 (1.00, 0)	0	0.555
Stroop Color-Word Test (% correct)	100 (100, 95.00)	100 (100, 97.50)	0.503
Stroop Color-Word Test (RT sec)	1.55 (1.91, 1.30)	1.30 (1.5, 1.25)	0.170
Tower of Hanoi-Sevilla (total time in sec)	137.27 (233.08, 67.15)	109.29 (149.82, 64.58)	0.308
Tower of Hanoi-Sevilla (correct movements)	10 (15.25, 8.25)	12.00 (13.00, 9.00)	0.914
Tower of Hanoi-Sevilla (errors)	4.00 (10.75, 2.00)	5.00 (8.50, 2.50)	0.682
Memory			
LMW-R			
True Recall Index	9.05 (9.40, 8.17)	9.40 (9.65, 8.95)	0.101
CI	5.28 (7.67, 2.86)	0	0.004
Memory gain	40.00 (49.15, 27.45)	30.00 (40.00, 30.00)	0.475
Self-Knowledge Index	0.15 (0.80, −0.27)	−0.20 (0.05, −0.71)	0.320
Index of Learning 1	0.33 (0.73, 0.09)	0.25 (0.45, 0)	0.249
Index of Learning 2	1.00 (1.69, 0.75)	1.00 (1.62, 0.50)	0.779
Index of Learning 3	1.25 (2.23, 0.85)	1.00 (1.95, 0.75)	0.559
Primacy effect	0.33 (0.86, −0.25)	0.50 (0.75, 0.08)	0.846
Recency effect	0.08 (0.62, −0.29)	0.44 (1.08, 0.33)	0.650
Consolidation Index	95.00 (100, 80)	100.00 (100.00, 95.00)	0.249
Digit Span Test (forward)	5.00 (7.00, 4.00)	6.00 (8.00, 5.00)	0.156
Digit Span Test (backward)	3.00 (4.5, 2.50)	5.00 (6.00, 4.00)	0.007
Complex Figure Test delayed score	18.75 (23.87, 8.37)	26.50 (33.25, 26.5)	<0.001
Visuoconstructive abilities			
Complex Figure Test copy score	36.00 (36.00, 35.00)	36 (36.00, 34.00)	0.914
Verbal fluency			
COWAT (phonological)	11.83 (13.66, 7.66)	12.66 (15.50, 10.33)	0.232
COWAT (semantic)	17.00 (19.41, 14.41)	17.33 (18.83, 15.50)	0.589

Data are expressed as median (75th and 25th percentiles) and *P* values are statistical comparisons between groups (Mann-Whitney nonparametric test). COWAT, Control Oral Word Association Test; RT, response time.

dition comparison = 3.38). In LMW-R, 2 × 2 ANOVA showed significant interaction between groups (acromegaly *vs.* healthy) and condition (immediate *vs.* delayed) (P = 0.02) (Fig. 1). Acromegaly patients had poorer verbal delayed recall (delayed-immediate mean difference = 0.625) than healthy controls (delayed-immediate mean difference = 0.308). Normative data comparison showed that patients' performance in the memory tasks fell below that of controls. In the Complex Figure Test, patients' median percentile was 35, whereas controls averaged 85 (seven patients and no controls performed below the 15th percentile). The median percentile for patients in the backward digit span test was 50; the control group's median was 25 (six patients and one control scored below the 15th percentile). In the delayed condition of LMW-R, patients' median percentile was 67.5, whereas the controls' was 85 (five patients and two controls scored below the 15th percentile). Normative data on the CI for the LMW-R comparison were not available.

A significant negative correlation was found between the patient group's GH and IGF-I levels and their Complex Figure Test memory score (GH, Spearman's $\rho = -0.51$, $P = 0.04$; IGF-I, $\rho = -0.54$, $P = 0.03$; IGF-I SD, $\rho = -0.67$, $P = 0.004$), implying that the higher the GH/IGF-I level, the poorer the task performance. Correlations between GH/IGF-I and performance in LMW-R's delayed verbal recall were not significant for IGF-I ($P = 0.15$) or IGF-I SD ($P = 0.13$), but a trend toward significance was detected for GH levels ($\rho = 0.44$, $P = 0.09$). This correlation indicates that the higher the GH levels, the worse the delayed

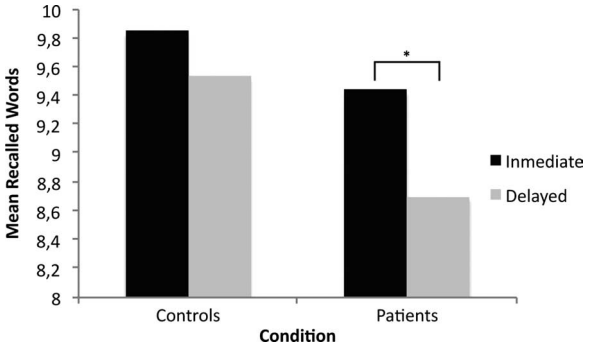

FIG. 1. Mean number of words recalled for both immediate and delayed condition. Control subjects' performances did not differ between conditions (mean difference between both conditions = 0.308; $P = 0.30$), whereas patients' performances were poorer in the delayed condition (mean difference = 0.625; $P = 0.02$). *, $P < 0.05$.

verbal recall. No significant correlations were detected between GH/IGF-I and other neuropsychological measures that differed between groups (all $P > 0.18$). There was no correlation between prolactin levels and neuropsychological performance (all $P > 0.43$).

Depression and QoL results

Patients with acromegaly showed higher depression scores in the BDI-II than healthy controls (median acromegaly group = 10.5; median healthy controls = 4; $P = 0.012$). Using BDI-II cutoffs, depression symptoms were found in 62.5% of the patients (50% showed mild depression, 12.6% moderate depression), and severe depression was not detected. In the control group, 15.4% showed mild depression symptoms. A significant relationship was detected between the presence of depression symptoms and group variables (Pearson's $\chi^2 = 0.56$; $P = 0.01$). However, BDI-II scores did not correlate significantly with significant neuropsychological measures or hormone levels (all $P > 0.18$). This lack of association between BDI-II scores and neuropsychological tests was also observed in the control group (all $P > 0.15$). In the AcroQoL, patients had a median score of 50.06. This measure correlated significantly with GH ($\rho = 0.54$; $P = 0.03$) and IGF-I ($\rho = 0.59$; $P = 0.02$). Moreover, a significant negative correlation was found between QoL and depression measures ($\rho = -0.56$; $P = 0.02$). A positive correlation was observed between AcroQoL and the Complex Figure Test memory score, which tended toward significance ($\rho = 0.47$; $P = 0.051$).

QEEG results

The spectral analysis showed significant differences in EEG components between groups. These differences were specific to parietal-occipital locations. Patients with acromegaly showed decreased activity in 10–11 Hz (both $P < 0.01$), 22–25 Hz (all $P < 0.05$), and 27–40 Hz (all $P < 0.05$). When EEG data were grouped into classical frequency bands, acromegaly patients showed less activity than healthy controls in α (8–12 Hz; $P = 0.03$), $\beta 3$ (22–30 Hz; $P = 0.04$), and γ (30–40 Hz; $P = 0.01$). Significant positive correlations were detected between the patient group's digit span performance and parietooccipital EEG activity (α, $\rho = 0.46$, $P = 0.02$; $\beta 3$, $\rho = 0.43$, $P = 0.03$; and γ, $\rho = 0.44$, $P = 0.02$). A significant positive correlation was also found between performance in the Complex Figure Test memory and γ activity ($\rho = 0.45$; $P = 0.02$). The remaining correlations did not reach significance (all $P > 0.17$). Figure 2 displays power spectra averaged across scalp regions for both clinical groups. F2

LORETA EEG source localization

A LORETA analysis was conducted to locate EEG cortical sources that differed between groups. This procedure was computed for each EEG frequency (from 1 to 40 Hz) and each frequency band. The results of the between-group comparisons (patients-controls) are represented in three orthogonal images that reflect regions of maximal statistical differences (Fig. 3). This analysis yielded significant differences between source magnitudes in the 9- to 12-Hz range (all $P < 0.01$), the 21- to 25-Hz range (P ranging from 0.15 to 0.43), and the 35- to 40-Hz range (P ranging from 0.003 to 0.043). Frequency band grouping analysis showed significant differences in α, $\beta 2$, $\beta 3$, and γ. In α, maximal differences were found in right inferior frontal lobe (maximal t value = -3.68; $P < 0.005$). Maximal differences were found in the right dorsolateral prefrontal cortex (DLPFC) in $\beta 2$ (maximal t value = -2.79; $P < 0.05$) and in the right parahippocampal cortex (maximal t value = -3.62; $P < 0.01$) in $\beta 3$. Patients also showed significantly less bilateral γ activity in DLPFC, showing maximal source difference in the left hemisphere (maximal t value = -3.47; $P < 0.05$). Figure 4 shows significant differences in LORETA analyses regardless of frequency band. Table 3 shows correlation analyses for LORETA and cognitive performance. F3 F4 T3

Discussion

Our study obtained two important results. First, patients with acromegaly showed poorer cognitive performance than healthy controls, particularly in memory tests. Second, neurophysiological assessment showed significant EEG power attenuation, namely decreased oscillatory activity in fast EEG bands (α, β, and γ) in prefrontal and middle temporal lobes. Decreased frontotemporal activity and impaired declarative memory functioning provide solid evidence of a concordance between electrophysio-

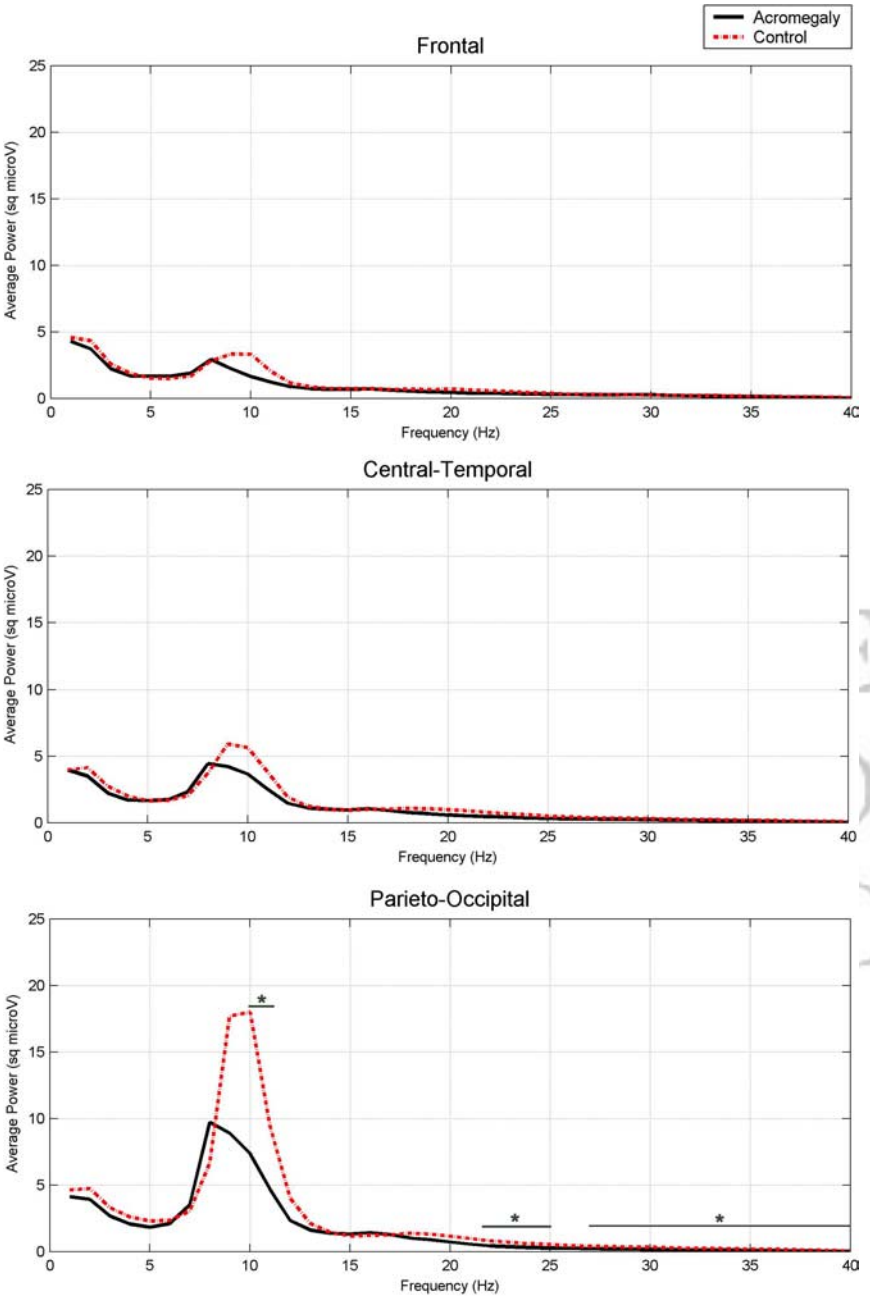

FIG. 2. Regional EEG power spectra comparing acromegaly patients (*black lines*) and healthy controls (*red dashed lines*). Overall, patients with acromegaly show less EEG power than matched controls, especially in the 8- to 25-Hz range. Statistical analyses show significant differences in α (8–10 Hz), $\beta 3$ (22–30 Hz), and γ (30–40 Hz) bands in parieto-occipital electrodes. *, $P < 0.05$.

logical and neuropsychological impairment. A distributed cortical network, encompassing prefrontal cortex and middle temporal structures, is essential for normal human memory function (30).

To the best of our knowledge, this study provides the first evidence of specific neurocognitive impairment in patients with active nontreated acromegaly. In addition, it identifies memory functions that appear to be altered: working memory, learning, and recall processes. Nevertheless, earlier studies cast doubt on the presence of memory impairment in patients with acromegaly. Richert *et al.* (31), using intelligence scales and subjective questionnaires on a sample of patients with active acromegaly, failed to find impairment in higher cognitive functions. This lack of cognitive impairment has also been postulated by Pantanetti *et al.* (32). Others (14), however, suggest that intelligence scales and subjective questionnaires may not be sensitive to subtler neurocognitive disorders. Our study is the first to identify these disorders using sensitive assessment tools.

Acromegaly patients showed difficulty in working memory tasks. Their performance, although normal in the forward digit span test, was worse in the backward digit span test. The backward test requires good memory span as well as preserved executive functions for processing control. Patients in our study did not show deficits in their verbal short-term memory, as demonstrated by results in the forward digit test. However, their results were worse in the backward digit test, where information was manipulated (executive control) in short-term memory. Neuroimaging studies have shown that the backward digit task activates brain regions that the forward task does not, namely mid-DLPFC regions (33). Several studies have shown that this task is useful for executive control assessment in frontal lobe surgery (34), frontal stroke syndromes (35), and frontotemporal dementia (36). In line with these findings, our study found that frontal lobe functioning was linked to backward digit task performance in acromegaly patients.

In addition, acromegaly patients did not display a shorter memory span during Luria's Word-Learning Task, compared with controls. However, a significant difference was found between patients and controls in this task. Patients with acromegaly showed more invented material in their free recalls because patients filled in or replaced the void left by non-recalled items with invented ones (16). This excessive memory contamination is seldom found in healthy populations, but it is not uncommon in neurological conditions affecting frontal lobes (37). A combination of memory and executive deficits seems to underpin memory contamination (38).

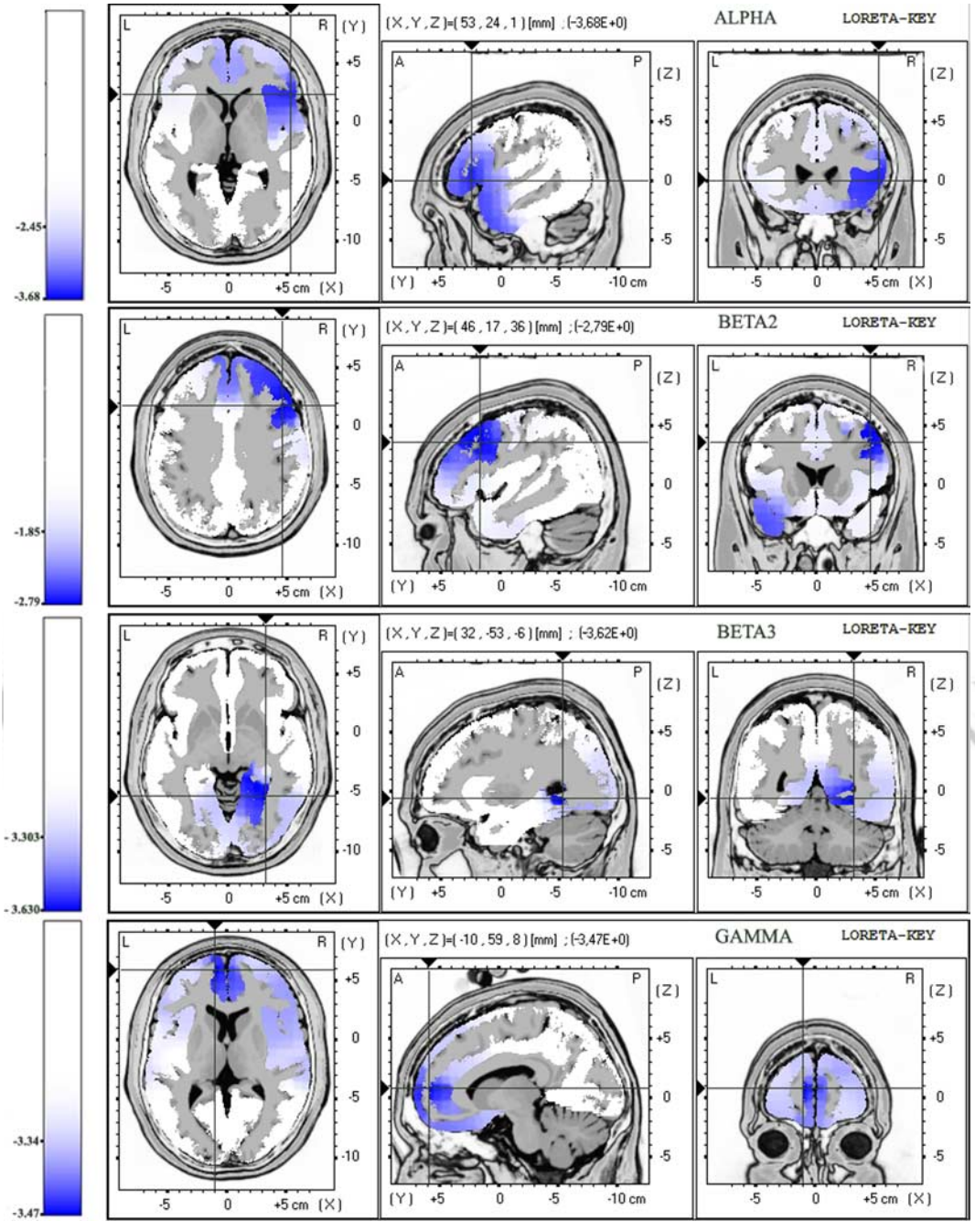

FIG. 3. LORETA comparisons between acromegaly group and healthy controls. Note that data show comparisons between group averages. Scales show maximal *t* value and minimal significant *t* value ($P < 0.05$). In acromegaly patients, areas with significantly decreased current (*blue*) in α, $\beta 2$, $\beta 3$, and γ bands include right inferior frontal lobe (α), right DLPFC ($\beta 2$), right parahippocampal cortex ($\beta 3$), and dorsolateral frontal lobe bilaterally (γ). *Vertical and horizontal line* intersection indicates maximal significant *t* value location.

Patients also performed worse in the two declarative memory measures: delayed recall and the Complex Figure Test memory condition. Faulty memory consolidation processes could be responsible for this deficit. Dudai (39) suggests that a consolidation process known as synaptic consolidation occurs after learning, where hippocampus and adjacent cortices in the middle temporal cortex play an essential role. In our study, the time elapsed between learning and delayed recall suggests that subject assessment took place at the initial stage of synaptic consolidation. Nevertheless, verbal delayed recall in our patients was not linked to middle temporal functioning, but rather to β activity in the right DLPFC cortex. Activation of right DLPFC is consistently observed in episodic memory retrieval neuroimaging studies (40, 41). This area could be responsible for implementing executive processes in memory (41).

Similarly, patients' performance was severely impaired in the Complex Figure Test visual memory condition,

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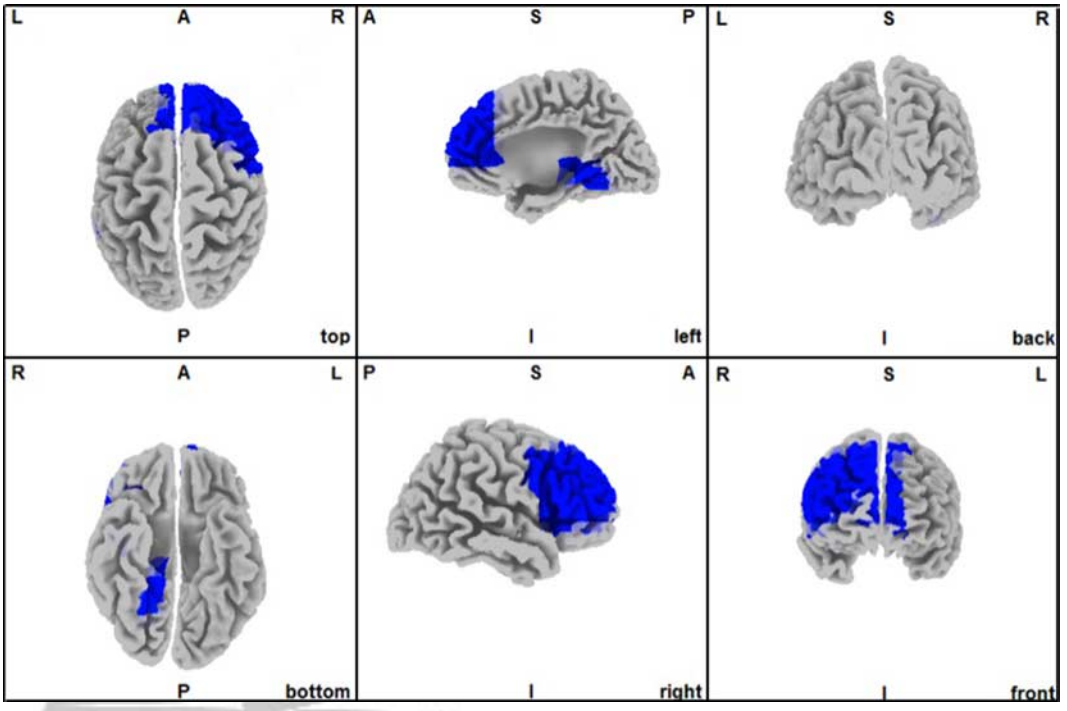

FIG. 4. Three-dimensional representation of LORETA differences regardless of frequency bands. Differences (acromegaly patients minus healthy controls) were plotted onto a three-dimensional brain surface, showing significantly decreased electrophysiological activity in a number of cortical regions including bilateral DLPFC, right medial prefrontal cortex, and right medial temporal lobe (see Fig. 3 for more details). L, Left; R, right; A, anterior; P, posterior; S, superior; I, inferior.

whereas no impairment was observed in the practice phase (copy condition) of this task. Visual memory impairment seems to underlie these findings. Performance in this task correlated positively with $\beta 3$ activity in the parahippocampal cortex. Human hippocampal formation plays a major role in memory for the Complex Figure Test (42). Moreover, hippocampal atrophy has been associated with impaired retention and recall in this task (43). Impairment in hippocampal-dependent memory may underlie these deficits in acromegaly patients.

The human hippocampus and adjacent areas (entorhinal, perirhinal, and parahippocampal regions) are mainly concerned with memory. Their widespread and reciprocal connections with the neocortex are essential factors in establishing long-term memory. As proposed by other authors (8, 9, 44), prolonged overexposure to endogenous supraphysiological levels of GH/IGF-I can cause neurophysiological or neuroanatomical changes, and brain areas with more binding sites to somatotropic axis hormones may be more vulnerable to this overexposure. In line with this hypothesis, our data show a strong negative correlation between GH/IGF-I levels and hippocampal-dependent memory performance. Tanriverdi *et al.* (8) found that high levels of GH/IGF-I are associated with impaired event-related P300 potentials, a neurophysiological measure sensitive to cognitive dysfunction. These results suggest a possible association between GH/IGF-I levels and severity of memory impairment. They also suggest the possibly deleterious neurophysiological effect of GH/IGF-I exposure on acromegaly and memory. More studies are needed to challenge this hypothesis.

QoL assessment in acromegaly has been studied extensively, highlighting the negative impact this disease has on a patient's well-being (45). In our study, GH/IGF-I levels

TABLE 3. *Post hoc* nonparametric correlations between LORETA cortical sources that showed less significant EEG activity and patients' performance in the four significant neuropsychological tasks

	α at right inferior frontal lobe	$\beta 2$ at right DLPFC	$\beta 3$ at right parahippocampal cortex	γ at left DLPFC
Complex Figure Test (memory)	0.03 (0.92)	−0.02 (0.96)	**0.70 (0.004)**	−0.14 (0.62)
LMW-R CI	−0.29 (0.29)	−0.22 (0.43)	−0.25 (0.36)	−0.23 (0.41)
Digit Span Test (backward)	−0.01 (0.99)	0.29 (0.29)	0.41 (0.13)	**0.72 (0.003)**
LMW-R delayed recall	0.08 (0.77)	**0.70 (0.005)**	0.43 (0.11)	0.04 (0.88)

Spearman's ρ is shown along with associated *P* value (in *parentheses*). Significant correlations appear in *bold*.

correlated strongly with QoL. However, other studies found no such correlations, although some reported weak correlations with morphological and physical changes (46). We also found a strong negative correlation between QoL and depression, whereby patients with more depression symptoms reported worse QoL. These findings suggest that depression may have a confounding role in the relationship between hormones and QoL.

Patients with acromegaly reported more depression symptoms compared with healthy subjects. According to the literature (11), most affective disorders in acromegaly are linked to major depressive episodes. This prevalence cannot be applied to our study sample, which lacked patients diagnosed with major depressive disorders. In fact, no patient in our sample reached the BDI-II cutoffs for severe depression. Our data on depression are more comparable to the report of Richert *et al.* (31), which focused on untreated acromegaly patients. Our results could be influenced by a number of factors, namely that our patients were newly diagnosed with acromegaly, they were younger than other patients found in the literature, and our depression questionnaire emphasized physical symptoms, which in fact could be due to the disease itself (47).

EEG power spectrum showed that acromegaly patients had decreased fast electrophysiological activity when compared with controls (reflected as less power in α, β, and γ bands during resting state). Fast EEG activity reflects the neurophysiological integrity of the cerebral cortex (48). Attenuation of fast EEG activity has also been associated with certain neurological conditions, such as Alzheimer's disease (49) or traumatic brain injury (50).

EEG source reconstruction using LORETA procedures revealed significantly decreased fast activity (α, $\beta 2$, $\beta 3$, and γ) in prefrontal and middle temporal cortices. In the healthy brain, these areas support certain cognitive functions, namely executive functions and memory (30, 51). Interestingly, activity in these brain areas was linked to cognitive performance in our patient sample. No correlation was found between right prefrontal α and $\beta 2$ bands and cognitive performance. The small sample size used in our study could be responsible for this lack of association. Our data suggest that certain cortical areas, which support short- and long-term memory processes, may be altered in acromegaly patients.

A number of factors could explain these results. First, chronic high levels of GH and IGF-I may have a deleterious effect on the central nervous system. For example, an impaired hypothalamic dopamine turnover rate was observed in studies using mouse models of acromegaly (44). Second, the high prevalence of vascular and metabolic complications found in acromegaly (1) could increase the risk of neurovascular disease. Sievers *et al.* (9) found a higher prevalence of white matter brain lesions in patients with hypertension and cardiovascular complications. Other potential modifiers of cognitive functions include the use of different drugs to treat acromegaly comorbidities (β-blockers or other hypotensive drugs) or the presence of pituitary deficiencies. Finally, psycho(patho)logical factors deriving from a chronic disease could also modify cognitive skills. However, depression symptoms were not associated with cognitive performance in our study. The relationship found in our study between cognitive performance and neurophysiological activity suggests an organic basis for cognitive deficits.

In conclusion, this is the first time that specific cognitive impairments, characterized by short- and long-term memory deficits, have been identified in patients with active acromegaly. Our findings suggest that high values of GH and IGF-I are related to the severity of these deficits. In addition, patients with acromegaly show less activation in brain areas strongly associated with memory, namely prefrontal and middle temporal cortices. The association between neurophysiological results and memory impairment found in our study strengthens the importance of these findings. What causes cognitive disorders in patients with acromegaly, and what is their clinical significance? Further testing, involving blood tests and brain imaging, may be needed to determine the causes of these findings and to demonstrate their significance when compared with healthy controls. This is particularly true if impairments improve after GH and IGF-I normalization through surgery and/or pharmacological treatment.

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