Clinical use of pre- and postsurgical evaluation of abnormal GH responses in acromegaly

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The criteria by which acromegalic patients are considered “cured” after surgical therapy are still controversial. Since the abnormal growth hormone (GH) increase after the administration of some agents has been demonstrated to be characteristic of the tumoral somatotrophs, its disappearance after surgery may be taken as an index of the complete removal of the tumor. Serum GH increases after thyrotropin-releasing hormone (TRH, 200 µg intravenously), gonadotropin-releasing hormone (Gn-RH, 100 µg intravenously), and sulpiride (100 mg intramuscularly) injected during dopamine infusion (DA-Slp test), were evaluated in 68 acromegalic patients before and after transnasosphenoidal adenomectomy, and every 12 to 18 months during a follow-up period of 6 months to 11 years (average 42 months). Forty-two patients had abnormal responses to at least one test before surgery: 32 out of 68 (47%) to TRH, six out of 40 (15%) to Gn-RH, and 20 out of 28 (71%) to the DA-Slp test. Of 18 patients who underwent all three tests, 78% had abnormal responses to at least one of them. Twenty-three patients became unresponsive after surgery, and none of them had a recurrence or became abnormally responsive again during the follow-up period. Three out of six patients with postoperative serum GH levels between 5.1 and 10 ng/ml and three out of six patients with postoperative serum GH levels between 2.1 and 5 ng/ml remained abnormally responsive: one of them relapsed 1 year after the operation. The abnormal responses were lost in all 11 patients whose postoperative serum GH levels were below 2 ng/ml, and abnormal responses were maintained in all the patients in whom surgery was considered unsuccessful because postoperative serum GH levels were higher than 10 ng/ml. The TRH, Gn-RH, and DA-Slp tests should thus be considered useful tools in verifying the total removal of an adenoma. The reappearance of active acromegaly in the patient with low postoperative GH levels, who was still responsive to TRH, should be regarded as a reactivation and not a true recurrence of the disease.

Key Words • acromegaly • human growth hormone • pituitary tumor • thyrotropin-releasing hormone • luteinizing hormone-releasing hormone • dopamine-sulpiride

Up to now, surgery is the most valid and widely used method of treating acromegaly. However, the criteria by which patients may be considered cured are still controversial. Although there is general agreement that the disease is still active in patients with postoperative serum growth hormone (GH) levels higher than 10 ng/ml, a question arises about levels lower than 10 ng/ml. In fact, such hormonal concentrations do not indicate that the tumor tissue is fully removed, since GH levels even lower than 5 ng/ml can be found in patients with documented active acromegaly. Also the recovery of normal responses to some dynamic tests cannot be taken as an index of the total eradication of the adenoma. In fact, the restoration of normal responses to insulin, arginine, and L-dopa has been described in patients whose persistently elevated GH levels suggested the presence of remnants of adenomatous tissue. Not even GH suppression after an oral glucose tolerance test (OGTT) may be considered a valid tool, as the interpretation of OGTT is difficult in patients with low serum GH levels, and some patients with active acromegaly show a normal inhibition of serum GH.

The disappearance of GH increase after the administration of thyrotropin-releasing hormone (TRH) has been reported to be useful in verifying whether some tumor tissue was left after surgery. In fact, this abnormal response occurs in acromegaly because of a direct action of TRH on the adenomatous somatotrophs, since it can be reproduced in vitro in tumor
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tissue but not in normal hypophyseal tissue. Similarly to TRH, gonadotropin-releasing hormone (Gn-RH) administration and sulpiride injection during dopamine infusion (DA-Slp test) have been demonstrated to induce GH elevations in quite a large proportion of acromegalic patients. These abnormal responses disappear after successful removal of the adenoma.

The role of these dynamic tests in the evaluation of surgical outcome in a series of 68 surgical patients followed over an 11-year span has been studied.

Clinical Material and Methods

Sixty-eight acromegalic patients (24 males and 44 females, aged 20 to 73 years) who underwent trans-sphenoidal surgery between 1971 and 1981 were studied before and after the surgical procedure. All had overt signs of active acromegaly. Basal serum GH levels ranged between 4.5 and 420 ng/ml (mean ± SE of 58.1 ± 8.9 ng/ml). Serum prolactin levels were determined in 56 cases and were found to be abnormally high in 21 (23 to 263 ng/ml in seven males and 28 to 64 ng/ml in 14 females). Radiological assessment of the sellar morphology included in all cases plain, frontal, and lateral skull radiographs, as well as tomograms obtained at 2-mm intervals by linear unidirectional motion. Pneumoencephalography and/or computerized tomography (CT) scans were also performed in the majority of patients. Modifications of the sellar morphology were classified according to Vezina and Maltais. A Type I sella was found in seven cases, a Type II in 32 cases, a Type III in 24 cases, and a Type IV in the remaining five cases.

Five patients had received cobalt-60 (Co) therapy 1 to 8 years before operation, and one patient also had yttrium-90 implants 6 and 4 years before surgery. At surgery, a pituitary tumor was found in all patients. Fifteen tumors were microadenomas, 55 were intrasellar adenomas, and 13 were macroadenomas with suprasellar expansion.

Serum GH modifications were studied after the following procedures. A 200-μg TRH injection was administered intravenously, and blood samples were taken 30 minutes before, and 0, 20, 30, and 60 minutes after the injection. An injection of Gn-RH (100 μg) was administered intravenously, and blood samples were taken 30 minutes before, and 0, 20, 30, and 60 minutes after the injection. In both tests, patients were considered responders when a serum GH increase of at least 50% and higher than 6 ng/ml above the value at 120 minutes was found after administration of sulpiride. This test was introduced in 1978, and was performed in 28 normotensive patients. Due to the possible hypertensive effect of dopamine, four other patients were excluded because of severe hypertension.

The TRH test was carried out in all 68 patients before and within 3 months after surgery, the Gn-RH test in 40, and the DA-Slp test in 28 patients. Subsequently, all responsive subjects were retested every 12 to 18 months. The mean follow-up period was 42 months, ranging between 6 months and 11 years. Twenty-four cases (35%) had a follow-up period longer than 4 years.

In addition, the L-dopa test (500 mg by mouth, with blood samples taken every 30 minutes for 3 hours) and insulin tolerance test (ITT, 0.1 to 0.2 IU/kg body weight, with blood samples taken every 30 minutes for 2 hours) were carried out in the 23 patients who became unresponsive to the tests described above. The L-Dopa test and ITT were designed to evaluate the ability of the residual pituitary tissue to secrete GH.

In all patients, the endocrine status was evaluated by basal serum triiodothyronine (T3), thyroxine (T4), cortisol, 17 beta-estradiol, and testosterone determinations. Pituitary reserve was studied before and after surgery in response to appropriate stimuli (TRH for thyroid-stimulating hormone (TSH); Gn-RH for gonadotropins; ITT and/or metyrapone for adrenocorticotropic hormone (ACTH)).

Before surgery, 43% of patients showed an impaired gonadotropin reserve, 24% had reduced ACTH reserve, and 40% showed an impaired or absent TSH response to TRH; three patients needed thyroid hormone and three needed cortisone replacement therapy. After surgery, impaired secretion of gonadotropins, ACTH, and TSH was found in 28%, 31%, and 42% of the cases, respectively; thyroid hormone and/or cortisone replacement therapy was necessary in eight and 12 patients, respectively.

Serum GH was evaluated by a double-antibody radioimmunoassay (RIA), using a specific antiserum raised in rabbit,* and the MRC 66/217 for labeling and standard curves. The coefficient of variation was 4.5% intra-assay and 7.9% inter-assay. Other hormonal determinations were performed by specific RIA methods. Statistical analyses were carried out by Student's t-test for unpaired data and by the Kolmogorov-Smirnov test, as appropriate.

Results

Before Surgery

Serum GH increased in 32 of the 68 patients (47%) after TRH, in six of the 40 (15%) after Gn-RH, and in 20 of the 28 (71%) after DA-Slp testing. Among the 18 patients who had all three tests performed, 14 (78%)

* Antiserum provided by Dow-Lepeit, Milan, Italy.
TABLE 1

Abnormal GH responses to TRH, Gn-RH, and DA-Slp tests in untreated acromegalic patients*

<table>
<thead>
<tr>
<th>Tests Performed</th>
<th>No. of Cases</th>
<th>Responders</th>
<th>No.</th>
<th>Percent</th>
<th>GH%†</th>
</tr>
</thead>
<tbody>
<tr>
<td>TRH</td>
<td>68</td>
<td>32</td>
<td>47</td>
<td></td>
<td>360 ± 83</td>
</tr>
<tr>
<td>Gn-RH</td>
<td>40</td>
<td>6</td>
<td>15</td>
<td></td>
<td>262 ± 64</td>
</tr>
<tr>
<td>DA-Slp</td>
<td>28</td>
<td>20</td>
<td>71</td>
<td></td>
<td>232 ± 71</td>
</tr>
<tr>
<td>TRH, Gn-RH, &amp; DA-Slp</td>
<td>18</td>
<td>14½</td>
<td>78</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Abbreviations: GH = human growth hormone; TRH = thyrotropin-releasing hormone; Gn-RH = gonadotropin-releasing hormone; DA-Slp = sulpiride injection during dopamine infusion.
† Mean percent of GH increases with respect to the basal levels ± standard error of the mean.
‡ Responders to at least one test.

were responsive to at least one test. For details see Tables 1 and 2. As far as GH basal levels or sellar morphology are concerned, no differences were seen between the groups of responders and non-responders (Table 3).

After Surgery

Basal serum GH dropped to less than 2 ng/ml in 18 patients, between 2.1 and 5 ng/ml in 17, between 5.1 and 10 ng/ml in 12, and still remained above 10 ng/ml in the remaining 21 cases. Comparing the results in the groups of responders and non-responders, the percentage of patients in whom GH concentrations decreased to less than 2 ng/ml was similar (26.1% versus 26.9%).

As for the postoperative modifications in GH secretion during dynamic tests, abnormal responses persisted in all 13 responsive patients in whom postoperative GH levels did not fall under 10 ng/ml, independently of the magnitude of the decrease. Abnormal responses disappeared in patients whose postoperative basal GH levels were below 2 ng/ml (11 patients). In 12 patients the basal GH decreased to levels between 2.1 and 5 ng/ml; in nine patients the abnormal responses disappeared and in three they persisted. In six patients the basal GH decreased to levels between 5.1 and 10 ng/ml: in three patients the abnormal responses disappeared, and in three they persisted. After surgery, patients who responded abnormally to two tests showed either a complete disappearance of both responses or the persistence of abnormal increases to the same tests as before (Fig. 1: in no case was only one of the two abnormal responses lost. Of the 29 responders (before surgery) whose postoperative GH basal levels dropped below 10 ng/ml, six maintained abnormal responses: three to TRH, one to the DA-Slp test, and two to both these tests. After surgery, 14 showed normal GH increase after the L-dopa test or ITT (data not shown).

After surgery, the pituitary response of tropic hor-

TABLE 2

Overall distribution of abnormal responses in the subgroup of responsive patients*

<table>
<thead>
<tr>
<th>Tests Performed</th>
<th>Responsive Cases</th>
<th>No. of Cases with Abnormal Responses to:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>TRH Only</td>
<td>Gn-RH Only</td>
</tr>
<tr>
<td>TRH</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>TRH &amp; Gn-RH</td>
<td>14</td>
<td>10</td>
</tr>
<tr>
<td>TRH &amp; DA-Slp</td>
<td>8</td>
<td>0</td>
</tr>
<tr>
<td>TRH, Gn-RH, &amp; DA-Slp</td>
<td>14</td>
<td>0</td>
</tr>
<tr>
<td>total cases</td>
<td>42</td>
<td>16</td>
</tr>
</tbody>
</table>

* Abbreviations: TRH = thyrotropin-releasing hormone; Gn-RH = gonadotropin-releasing hormone; DA-Slp = sulpiride injection during dopamine infusion; — = test not done.
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FIG. 2. Serum growth hormone (GH) levels in a patient responsive to thyrotropin-releasing hormone (TRH). Note the disappearance of GH response to TRH soon after surgery (1971 post) and during 11 years of follow-up review. Closed circles = before surgery; open circles = after surgery.

mones, other than GH, was normal in 17 of the 23 patients in whom the aberrant GH responses disappeared. The 14 patients in whom GH increased after the L-dopa test or ITT were all part of this group. Three of the remaining six patients had unitropic deficiencies (two TSH and one ACTH), one had gonadotropin and TSH deficits and one gonadotropin and ACTH deficiencies. The sixth patient had panhypopituitarism. Only the last two patients needed replacement therapy.

Follow-Up Study

In the group of unresponsive patients, those with postsurgical GH basal levels below 2 ng/ml did not show signs of recurrence, whereas one patient with serum GH between 2.1 and 5 ng/ml and four with GH between 5.1 and 10 ng/ml relapsed. In the group of responsive patients, no sign of recurrence or reappearance of abnormal responses was seen in any of the patients who became unresponsive after surgery over a mean period of 35 months (range 6 months to 11 years), irrespective of their GH basal levels (Fig. 2).

Of the 19 patients who were still responsive after surgery, 17 were available for follow-up review. Sixteen patients underwent 60Co radiotherapy and then received bromocriptine. In nine of these cases the pattern of abnormal GH responses to the tests, repeated every 12 to 18 months, remained unmodified, although basal serum GH levels gradually lowered in six patients (Fig. 3). Abnormal GH responses persisted as well during bromocriptine therapy, although a further reduction in GH levels down to 5 ng/ml was observed in three patients. In six other patients basal GH levels decreased below 5 ng/ml, and these patients lost abnormal responses 2 to 4 years after radiation therapy (Fig. 4).

The remaining patient whose mean basal GH levels were 3.8 ng/ml was still responsive to the TRH test. He relapsed within a year and underwent reoperation.

<table>
<thead>
<tr>
<th>Patient Group</th>
<th>Mean Basal GH (ng/ml ± SD)</th>
<th>Sella Type</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Preop</td>
<td>Postop</td>
</tr>
<tr>
<td>responders</td>
<td>52 ± 51.2</td>
<td>12.6 ± 18.6</td>
</tr>
<tr>
<td>nonresponders</td>
<td>55.4 ± 70</td>
<td>15.0 ± 19.1</td>
</tr>
</tbody>
</table>

* Sella type classified according to Vezina and Maltais. Abbreviations: GH = human growth hormone; TRH = thyrotropin-releasing hormone; Gn-RH = gonadotropin-releasing hormone; DA-SIp = sulpiride injection during dopamine infusion; NS = not significant; SD = standard deviation.

† Student's t-test for unpaired data.
‡ Kolmogorov-Smirnov test.
FIG. 4. Serum growth hormone (GH) levels in a patient responsive to sulpiride (Slp) injection during dopamine infusion (DA-Slp). The abnormal response was still present after surgery, but declined after irradiation. **Closed circles** = before surgery; **open circles** = after surgery; **triangles** = after irradiation.

**Discussion**

The data presented in this study demonstrate that 78% of acromegalic patients show abnormal responses to stimuli (TRH, Gn-RH, and DA-Slp tests) which are presumed to reflect alterations in the cellular membrane receptors and to be peculiar to the adenomatous somatotrophs. 2,7,9,16,25,26,35 In this view, the disappearance of such aberrant responses after surgery would indicate that tumor tissue is fully eradicated.

Forty-two patients out of 68 were found to be responsive to at least one test. Twenty-three patients became unresponsive after transsphenoidal selective adenomectomy, and none of them either had a recurrence or became responsive again during the follow-up period lasting 6 months to 11 years (mean 35 months). Three of the six patients with postoperative serum GH levels between 5.1 and 10 ng/ml and three of the 12 with postoperative serum GH values between 2.1 and 5 ng/ml remained responsive; one of them relapsed 1 year after the operation. The aberrant responses were lost in all patients whose postoperative serum GH levels were below 2 ng/ml, but were maintained in all the patients in whom surgery was considered unsuccessful because of high postoperative serum GH levels.

As for the TRH test, the present findings confirm our previous data 11,14 in a larger group of patients and during a longer follow-up period, and are in agreement with other recent reports. Arafah, et al., 1 and Schuster, et al., 32 observed recurrences in three out of seven and in two out of four patients, respectively, with low postsurgical levels of GH who were still responsive to TRH. D. K. Ludecke, et al. (unpublished data, 1980), also described recurrences in patients with positive TRH tests. One could argue that TRH is also able to induce serum GH increases in pathological conditions other than acromegaly, such as mental depression, 23 anorexia nervosa, 24 renal failure, 15 liver cirrhosis, 28 and hypothyroidism. 4 It is noteworthy that in such diseases the pattern of GH response to TRH is quite different from that seen in acromegaly: increases are smoother, usually delayed, and extremely variable. Such aberrant responses are therefore assumed to be related to metabolic abnormalities occurring in the above-mentioned diseases, and to their effects on the central nervous system. 7,15,28 On the contrary, GH increases observed in acromegaly are marked (mean increase of 360% in the present series), rapid (within a few minutes, peak at 20 to 30 minutes), and reproducible. Furthermore, they have been reproduced in vitro in GH-secreting pituitary tumors, but not in normal hypophyseal tissue. 7,10,25 The major limitation of the TRH test is that not all acromegalic patients are responsive; in the present study, only 32 of 68 patients (47%) showed increased GH after TRH before surgery. This incidence is in agreement with previous reports. 8,30

This finding led us to evaluate anomalous responses to two other tests sharing a pituitary level of action with TRH: the Gn-RH and DA-Slp tests. Both were demonstrated to stimulate GH secretion in cases of active acromegaly, but not in normal subjects. 2,8,10,31 The present data confirm that these abnormal responses, as the one provoked by TRH, disappear in successfully treated patients, and can be assumed to be an index of the total removal of the adenoma. This concept is emphasized by the finding that the abnormal responses, present in the same patient, behaved in the same manner after surgery (Fig. 1). Moreover, the patterns of abnormal GH increases do not change during the follow-up study in unsuccessfully treated patients, thus confirming the reproducibility of the tests. On the other hand, a progressive decline in GH peaks, which paralleled the lowering of basal GH levels, was noticed in patients who underwent radiotherapy successfully. The incidence of positive responses to the DA-Slp test was higher than that of the responses to TRH (71% versus 47%). On the contrary, Gn-RH elicited a GH increase in few patients (six of 40, or 15%); however, it is noteworthy that three patients showed only this abnormal response. By using all three tests, 78% of acromegalic patients could be monitored by the evaluation of abnormal GH responses.

The finding that GH basal levels, the sella type (classified according to Vezina and Maltais 36), and the GH decrease after surgery are similar in the responsive and unresponsive patients suggests that the abnormal response is not related to the severity of the disease. The
disappearance of abnormal GH responses after surgery might be attributed to either the selective ablation of all adenomatous somatotrophs or the complete removal of the hypophyseal gland. However, the finding of normal GH responses to the L-dopa test and/or ITT, and of a normal pituitary reserve of other tropins, documents the presence of normal pituitary cells in most of our patients.

The follow-up study of patients whose GH secretion became completely normal might throw light on the pathogenesis of acromegaly. It is a matter of debate whether acromegaly is a primary pituitary disease or a consequence of a defect in the hypothalamic regulation.5,6,8 If we hold the last hypothesis to be true, we should expect a recurrence of the disease in those patients who underwent a complete and selective adenectomy. In fact, the hypothalamic alteration would persist, thus leading to development of a new adenoma. None of our patients whose GH responses were normalized after surgery showed a recurrence. These patients were followed for up to 11 years, but a longer follow-up period might be useful. Data available in the literature about the recurrence of GH-secreting adenomas cannot be used for this goal. In fact the term “recurrence,” as quoted in the literature, may either signify a true recurrence or a reactivation of the disease due to incomplete removal of the adenoma.22 One of our patients with low postoperative GH levels but who was still responsive to TRH again showed clinical evidence of active acromegaly. This should be considered a reactivation of the disease rather than a recurrence. In our opinion, the use of dynamic tests together with a longer follow-up period could be an important step to elucidate the problem of recurrence. In fact, a late relapse of acromegaly in a patient with a normalized response pattern to these tests would most likely be a true recurrence, provided actual recurrences in acromegaly do exist.

Addendum

Soon after this paper was submitted, Thorner, et al., described a case of a patient with acromegaly due to a pancreatic islet tumor secreting a growth hormone (GH)-releasing factor (Thorner MO, Perryman RL, Cronin MJ, et al: Somatotroph hyperplasia. Successful treatment of acromegaly by removal of a pancreatic islet tumor secreting a growth hormone-releasing factor. J Clin Invest 70:965–977, 1982). Their patient showed an abnormal increase in GH after injection of thyrotropin-releasing hormone (TRH). The increase disappeared after removal of the tumor. This finding might challenge the current interpretation of the abnormal GH responses occurring in GH-secreting pituitary adenomas. However, as no in vitro study of TRH responsiveness of hyperplastic somatotrophs was carried out, the hypothesis that, in this rare condition, the GH increase after TRH was mediated by release of GH-releasing factor from the pancreatic tumor cannot be excluded.

References


Manuscript received October 12, 1982, Accepted in final form April 21, 1983.
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