ABSTRACT

Glucocorticoid-remediable aldosteronism (GRA) is a rare form of inherited hypertension caused by a characteristic gene duplication. With the advent of definitive genetic testing for GRA, the performance of the traditional screening test for GRA, the dexamethasone suppression test (DST), can be evaluated. We compared the DST to direct genetic testing in 24 patients referred for genetic screening for GRA (12 GRA positive and 12 GRA negative) based on clinical and biochemical findings, DST, and family history. Plasma aldosterone was measured before and after oral dexamethasone administration to determine the extent to which aldosterone was suppressed by glucocorticoids in each patient group. The results of the DST in these subjects were also compared to those in 19 historical patients with primary aldosteronism [4 bilateral hyperplasia and 15 aldosterone-producing adenoma (APA)] reported previously. The DST differentiates GRA-positive from GRA-negative patients with 92% sensitivity and 100% specificity. Cutoffs based on the post-DST plasma aldosterone level (<4 ng/dL) or percent suppression compared to baseline (>50%) were equally effective in correctly diagnosing GRA (only one GRA-positive patient would have been incorrectly diagnosed). However, DST in 15 APA patients revealed that 33% had greater than 80% suppression of aldosterone, and 1 had aldosterone levels below 4 ng/dL.

We conclude that a post-DST aldosterone level below 4 ng/dL will correctly diagnose GRA patients with high sensitivity and specificity. Suppression compared to baseline can be misleading, as evidenced by the results in APA patients and referred subjects who genetically screened negative. (J Clin Endocrinol Metab 82: 3570–3573, 1997)

Subjects and Methods

Subjects

Retrospective review of the records of patients that had been genetically screened for GRA by the International Registry for Glucocorticoid-Remediable Aldosteronism revealed 24 individuals that had prior DST with baseline and post-DST plasma aldosterone levels. These patients were referred for genetic testing based on the clinical setting, family history, or results of DST (3, 4). Twelve subjects tested positive (GRA+), and 12 tested negative (GRA−).

The results of DST in these patients were compared to results in patients with primary hyperaldosteronism reported previously [15 patients with aldosterone-producing adenomas (APA) (5, 6) and 4 patients with idiopathic hyperaldosteronism (IHA) (7)]. APA patients were diagnosed on the basis of biochemical testing alone (n = 2) or biochemical testing and adrenal venous sampling (n = 5), surgery (n = 6), or adrenal venography (n = 2). IHA was diagnosed on the basis of biochemical findings and adrenal scintigraphy (n = 3) or postmortem examination (n = 1). Genetic testing was not performed in these historical controls.

Genetic testing

Total genomic DNA was extracted from 10–20 mL peripheral venous blood as previously described (8). The chimeric 11β-hydroxylase/aldosterone synthase gene was identified using a previously reported Southern blot technique (4). Genetic testing was performed without prior knowledge of the response to DST.
DST

Most subjects underwent low dose DST based on that originally described by Liddle et al. (9, 10). Dexamethasone (0.75–2.0 mg/day) was administered orally for at least 2 days. Plasma aldosterone was measured before and after the DST.

Statistical analysis

The two patient groups were compared using an unpaired t test with an α level of 0.05. Results are expressed as the mean ± SEM. Sensitivity, specificity, positive predictive value, and negative predictive value were calculated for DST using genetic testing as the definitive test. Baseline and post-DST aldosterone levels and percent suppression of aldosterone were calculated for DST using genetic testing as the definitive test. Baseline and post-DST in these same subjects. After DST, GRA subjects showed a significantly greater percent suppression of aldosterone than did GRA subjects. *, P < 0.002; ‡, P < 0.0001.

Genetically screened subjects

The GRA+ subjects (n = 12) were similar to the GRA− subjects (n = 12) with respect to gender, systolic blood pressure, diastolic blood pressure, baseline serum aldosterone, and PRA (Table 1). GRA+ and GRA− subjects differed significantly with respect to age (mean age, 21.2 ± 3.6 and 36.4 ± 4.2 yr, respectively; P = 0.02). The median plasma aldosterone/PRA ratio for both groups was 141.5, consistent with a diagnosis of primary aldosteronism where the ratio usually exceeds 30 (11).

Both dose (mean, 1.7 ± 0.13 mg/day; range, 0.75–2 mg/day) and duration of dexamethasone treatment (mean, 4.5 ± 0.73 days; range, 1–15 days) varied widely for all subjects. Although the two groups did not differ significantly in terms of dexamethasone dose or duration, GRA− patients showed a trend toward a longer duration of DST (P = 0.06; Table 1).

Although all subjects had aldosterone measured during the DST, pre- and post-DST cortisol levels were measured in only 7 of 12 GRA+ patients and in 3 of 12 GRA− patients. Post-DST cortisol levels indicated that the dexamethasone dose was sufficient to suppress cortisol levels to less than 5 ng/dL in these subjects (data not shown). Of those subjects without post-DST cortisol measurements, all had been given dexamethasone at doses that have been previously shown to suppress morning cortisol levels in normal subjects.

Baseline aldosterone levels tended to be higher, but were not significantly different, in the GRA− group (54.2 ± 13.9 vs. 26.0 ± 3.6 ng/dL, respectively; P = 0.06; Fig. 1A). However, post-DST aldosterone levels were significantly lower in GRA− patients compared to GRA+ subjects (2.4 ± 0.44 and 22.0 ± 5.5 ng/dL, respectively; P < 0.0002). In addition, the percent suppression of aldosterone vs baseline was significantly greater in GRA+ subjects (88 ± 3% vs. 54 ± 4%, P < 0.0001; Fig. 1B).

Comparison of DST to genetic testing showed good discrimination between GRA+ and GRA− when either the post-DST plasma aldosterone (GRA+, <4 ng/dL) or the percent suppression of aldosterone vs. baseline (GRA+, >80% suppression) end points were employed. The sensitivity and specificity were, respectively, 91.7% and 100%, with positive and negative predictive values of 100% and 99.1% for both tests (the latter assumes a 20% prevalence of disease in our study population, which approximates that seen in patients referred for genetic testing). The use of either end point resulted in correct identification of all GRA− subjects and a misclassification of one GRA+ subject.
**Historical subjects with primary aldosteronism (5–7)**

Compared to our study subjects, the historical patients had been given a greater mean dose of dexamethasone (2.1 ± 0.2 mg/day; P = 0.02) and a shorter mean duration of treatment (2.2 ± 0.1 days; P < 0.01). DST in 15 APA patients resulted in mean baseline plasma aldosterone levels (50.0 ± 7.3 ng/dL) declining to 23.1 ± 5.2 ng/dL (49 ± 9% suppression). Basal and post-DST plasma aldosterone levels and percent suppression of plasma aldosterone in APA patients were significantly different from those in GRA subjects (P values 0.01, 0.02, and 0.001, respectively), but not from those in GRA subjects (Fig. 2).

A post-DST plasma aldosterone measurement below 4 ng/dL resulted in 14 of 15 APA patients being correctly classified as GRA−. However, only 10 of 15 APA patients would be correctly classified as GRA− if 80% suppression vs. baseline was used. The small group of 4 patients with IHA did not exhibit a significant reduction in aldosterone levels post-DST (pre-DST, 23.2 ± 3.9 ng/dL; post-DST, 24.9 ± 5.7; 0 ± 6% suppression). Table 2 summarizes the results of DST in all 4 patient groups.

**TABLE 2.** Results of dexamethasone suppression testing in 24 subjects genetically screened for the chimeric gene and 19 subjects with primary aldosteronism reported in the literature

<table>
<thead>
<tr>
<th>Patient Group</th>
<th>n</th>
<th>Pre-DST aldosterone (ng/dL)</th>
<th>Post-DST aldosterone (ng/dL)</th>
<th>% Suppression</th>
<th>DST accuracya (Aldosterone &lt;4 ng/dL)</th>
<th>DST accuracyb (Suppression &gt;80%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GRA+</td>
<td>12</td>
<td>26.0 ± 3.6</td>
<td>2.4 ± 0.44</td>
<td>88 ± 3</td>
<td>11/12</td>
<td>11/12</td>
</tr>
<tr>
<td>GRA−</td>
<td>12</td>
<td>54.2 ± 13.9</td>
<td>22.0 ± 5.5</td>
<td>54 ± 4</td>
<td>12/12</td>
<td>12/12</td>
</tr>
<tr>
<td>APAa</td>
<td>15</td>
<td>50.0 ± 7.3</td>
<td>23.1 ± 5.2</td>
<td>49 ± 9</td>
<td>14/15</td>
<td>16/15</td>
</tr>
<tr>
<td>IHA−</td>
<td>4</td>
<td>23.2 ± 3.9</td>
<td>24.9 ± 5.7</td>
<td>0 ± 6</td>
<td>N/A</td>
<td>N/A</td>
</tr>
</tbody>
</table>

GRA+, GRA positive; GRA−, GRA negative; APA, aldosterone-producing adenoma; IHA, idiopathic hyperaldosteronism; N/A, individual data not available.

a DST accuracy represents the number of subjects correctly classified after DST using the criteria of the post-DST aldosterone level or the percent aldosterone suppression.

b See Ref. 5 and 6.

c See Ref. 7.

**Discussion**

The identification of the genetic basis of GRA by Lifton et al. (4) and the recent development of a simple PCR test (12, 13) have permitted the diagnosis of GRA using direct genetic testing. However, many clinicians continue to perform a DST to determine who should be genetically screened or use it in lieu of genetic testing altogether. Although the DST has been used to diagnose GRA since the syndrome was first described by Sutherland et al. (14), there is no consensus as to the how the test should be performed. Doses ranging from 2–8 mg/day for 2 days to 4 weeks have been used (15–20). Finally, it appears that clinicians are uncertain how they should interpret the results of DST in the differential diagnosis of patients with biochemically established primary aldosteronism.

The present study represents a retrospective review of patients referred for genetic testing to the International Registry for Glucocorticoid-Remediable Aldosteronism. Of 24 subjects with pre- and post-DST aldosterone measurements (12 GRA positive and 12 GRA negative), the DST proved to be accurate in correctly diagnosing GRA if the proper suppression criteria were applied. Cut-offs based on absolute aldosterone levels (<4 ng/dL) or percent suppression of aldosterone (>80%) were equally effective in correctly diagnosing GRA, with only 1 subject that was GRA+ being misclassified. This subject’s plasma aldosterone level fell from 14.3 to 6.4 ng/dL (55% suppression) after the administration of 0.5 mg dexamethasone every 6 h for 3 days. However, treatment compliance could not be confirmed because cortisol was not measured post-DST. This underscores the importance of measuring both aldosterone and cortisol concurrently when performing DST to ensure that noncompliance with dexamethasone administration does not result in a false negative test.

In considering the differential diagnosis of GRA, there are a number of mineralocorticoid excess states in which the elevation in blood pressure may be glucocorticoid responsive (1). These include congenital and acquired 11β-hydroxysteroid dehydrogenase deficiency as well as congenital adrenal hyperplasia due to 11β-hydroxylase or 17α-hydroxylase deficiency. Although lowering blood pressure with DST in these syndromes is similar to that seen in GRA, these low renin syndromes can be distinguished because the mineralocorticoid excess state is caused by a steroid other than aldosterone (e.g. cortisone in 11β-hydroxysteroid deficiency). However, DST in patients with the more common etiologies of primary aldosteronism, such as APA and IHA, could cause...
As the final diagnosis was unknown in our subjects who genetically screened negative for GRA, we reviewed the literature for the plasma aldosterone response to DST in patients with primary aldosteronism. When the above DST diagnostic criteria were applied to 15 patients with APA (5, 6), the post-DST aldosterone level was more accurate than the percent suppression in correctly classifying these patients as GRA; 14 of 15 APA subjects had post-DST aldosterone levels above 4 ng/dL. On the other hand, one third of the APA patients had more than 80% suppression of aldosterone levels post-DST (5, 6). Thus, even though APA patients often show clear-cut dexamethasone suppressibility of aldosterone, the post-DST aldosterone level is usually higher than those seen in GRA patients.

Thus, DST can be used to correctly diagnose almost all patients with GRA if appropriate post-DST aldosterone criteria are applied. We believe that DST is best accomplished by administering 0.5 mg dexamethasone, orally, every 6 h for 2 days; concurrent measurement of cortisol with aldosterone is recommended to document adequate suppression of ACTH. Longer duration of DST (>1 week), as has been performed by some investigators (20, 25), is not recommended because this may result in reactivation of the renin-angiotensin system in GRA patients, possibly yielding false negative results. Although a cut-off of more than 80% suppression of aldosterone after DST would clearly have misclassified some patients with APA, a post-DST aldosterone level less than 4 ng/dL was seen in less than 7% of APA patients. Thus, there was overlap in the post-DST aldosterone levels in the GRA and APA groups. As the DST cannot definitively distinguish APA from GRA, direct genetic testing still appears to be the definitive confirmatory test (100% sensitivity and 100% specificity). This is underscored by a report of autonomous aldosterone production with failure to suppress after DST in an established GRA family (24).

When should the DST be administered? GRA is rare and has a low prevalence, even among patients with biochemical primary hyperaldosteronism. It is, therefore, important to stress that the routine use of the DST as part of the initial diagnostic evaluation of patients with primary hyperaldosteronism is inappropriate. Rather, we propose that the DST be used in patients with biochemical primary hyperaldosteronism, who have a suggestive clinical history (i.e. early-onset hypertension in the proband and in first degree relatives) and negative computed tomography imaging of the adrenals. Finally, a positive DST should not displace the primacy of direct genetic testing in the diagnosis of GRA.

The fall in aldosterone to nearly undetectable levels after DST in GRA is expected and reflects the sole control of aldosterone by ACTH in this disorder. The significant suppression of aldosterone levels after DST in APA reflects the well recognized regulation of aldosterone by ACTH in this disorder (16, 17, 24–26). However, autonomous production of aldosterone by the neoplasm accounts for the failure of aldosterone levels to fall to very low or nearly undetectable levels. It was with these observations in mind that the late Dr. Stanley Ulick favored the term GRA over dexamethasone-suppressible hyperaldosteronism for the disorder that he biochemically characterized by the measurement of unique steroid metabolites (6). These data underscore his penetrating contributions and insights.