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The Value of the Low-Dose Dexamethasone Suppression Test in the Differential Diagnosis of Hyperandrogenism in Women

GREGORY A. KALTSAS, ANDREA M. ISIDORI, BLERINA P. KOLA, ROB H. SKELLY, SHERN L. CHEW, PAUL J. JENKINS, JOHN P. MONSON, ASHLEY B. GROSSMAN, AND G. MICHAEL BESSER

Department of Endocrinology, St. Bartholomew's Hospital, London ECIA 7BE, United Kingdom

We studied 211 hyperandrogenic women with respect to clinical presentation, basal androgen levels, and the degree of cortisol and androgen suppression during a 48-h low-dose (2 mg) dexamethasone-suppression test (LDDST) to exclude ovarian and adrenal tumors. In 42 women with elevated testosterone levels, 21 of whom failed to suppress testosterone during the LDDST, the response of serum androgen levels during a 4-wk administration of 7.5 mg prednisolone in a reverse circadian regimen was also assessed. These results were compared with an additional 17 patients with histologically proven androgen-secreting tumors.

Clinical presentation alone was suggestive of a virilizing tumor in 70% of patients with tumors. Serum testosterone, although occasionally only marginally elevated, was the sole androgen that was elevated in every patient with a tumor. After the LDDST, none of the patients with tumors obtained either a greater than 40% reduction or normalization of the previously elevated testosterone levels, whereas 88% of patients with nontumorous hyperandrogenism showed either normalization or suppression of more than 40%. With one exception, all of the patients with nontumorous hyperandrogenism who showed inadequate suppression of testosterone during the LDDST, and were treated with prednisolone, normalized the previously elevated androgens after 1 month of administration.

In summary, in women presenting with hyperandrogenism, lack of testosterone suppression during the LDDST is associated with 100% sensitivity and 88% specificity in distinguishing patients with ovarian and adrenal androgen-secreting tumors from patients with nontumorous hyperandrogenism in this small series. The LDDST is an easy to perform screening test that can also identify causes of hyperandrogenism due to altered glucocorticoid secretion. (*J Clin Endocrinol Metab* 88: 2634–2643, 2003)

CLINICAL FEATURES SUGGESTIVE of hyperandrogenism (hirsutism, acne, alopecia, and menstrual irregularity) are a common problem in women, and have been linked to excessive androgen production from the ovaries or the adrenal glands or both (1). Although the majority of patients will be found to have nontumorous hyperandrogenism, it is important to identify the small proportion of patients with virilizing adrenal or ovarian tumors (2–4). The extent to which patients with hyperandrogenism should be investigated to exclude the likelihood of a tumor is still a matter of debate (2–5). It has been suggested that the rapid onset of symptoms and a single measurement of serum testosterone alone will suffice to identify patients who require more extensive investigation (3, 5). However, some tumors may run an indolent course with subtle symptoms resembling those of patients with nontumorous hyperandrogenism (2). In addition, a single testosterone measurement has a low predictive value for an androgen-secreting tumor because it overlaps with values obtained from women with nontumorous hyperandrogenism, and particularly because some tumors may secrete androgens other than testosterone (2, 6, 7). Therefore, discrimination between these disorders on the basis of symptoms or basal testosterone levels alone may not be possible.

Androgen suppression in response to glucocorticoid administration has been used previously to exclude patients with virilizing adrenal tumors and to identify the source of excessive androgen production in patients with nontumorous hyperandrogenism (2, 8–10); failure to achieve suppression of the elevated androgen levels is considered characteristic of a virilizing adrenal tumor, whereas good suppression is suggestive of a nonneoplastic adrenal source (2, 6, 8–10). Cortisol suppression in response to dexamethasone can also identify patients with Cushing's syndrome, as well as patients with either a partial form of glucocorticoid resistance or alterations of glucocorticoid synthesis and metabolism (2, 3, 11–13). However, although there is considerable evidence that dexamethasone suppression may be useful in the overall diagnosis of hyperandrogenism (2), descriptions of the ability of the dexamethasone suppression test to identify patients harboring ovarian tumors are limited to single case reports (6, 14–22).

For some years, we have been performing the 48-h low-dose dexamethasone-suppression test (LDDST) routinely in all women presenting with hyperandrogenism who had an elevation of at least one of their basal androgens to distinguish between the different causes of hyperandrogenism. In this study, we have analyzed the case records of all 211 patients with nontumorous hyperandrogenism who under-

Abbreviations: CT, Computed tomography; DHEAS, dehydroepiandrosterone sulfate; LDDST, low-dose dexamethasone-suppression test; 17-OH-PG, 17-hydroxyprogesterone; PCO, polycystic ovaries; PCOS, polycystic ovary syndrome.

went an LDDST over a period of 8 yr and compared these to 17 patients with histologically proven ovarian or adrenal androgen-secreting tumors.

Patients and Methods

Patients

We analyzed retrospectively the case notes of 211 consecutive women who presented over an 8-yr period (1990–1998) with symptoms of hyperandrogenism and who had undergone an LDDST. None of these patients had evidence of an androgen-secreting tumor at diagnosis or during the subsequent follow-up period of 2–11 yr (median, 6 yr). The results in these patients were compared with those of 17 patients with histologically proven androgen-secreting tumors. The patients with tumors were collected over a 20-yr period (1980–2000), and 11 of them underwent an LDDST. Symptoms and signs of hyperandrogenism were defined as previously published (23, 24). Hirsutism and acne were graded as absent, mild, moderate, and severe; alopecia as present or absent; menstrual pattern as normal (21–35 d), oligomenorrhea (35 d to 6 months), amenorrhea (>6 months), and polymenorrhea (<21 d). With the exception of those patients with adrenal tumors that cosecreted cortisol and androgens (n = 5; Table 1), patients with hyperandrogenism who also presented clinical features suggestive of Cushing's syndrome were not included in the study. The study was approved as an institutional case-note review subject to departmental authorization.

Hormonal determinations

All patients underwent evaluation for hyperandrogenism according to our established protocol (23). This included measurement of basal serum androgens at 0900 h [testosterone, androstenedione, dehydroepiandrosterone sulfate (DHEAS)], cortisol, and 17-hydroxyprogesterone (17-OH-PG). All plasma measurements were performed during the follicular phase of the menstrual cycle in patients who were menstruating, off any estrogenic medication. Patients who obtained a basal 17-OH-PG value greater than 7 nmol/liter (>2.3 ng/ml) underwent a standard short ACTH stimulation test to exclude late-onset congenital adrenal hyperplasia. The 48-h 2 mg/d LDDST was performed according to a standardized protocol (23–25): blood samples were collected at 0900 h before and after 8 doses of dexamethasone administered as 0.5 mg every 6 h for 48 h. The second blood sample was obtained 6 h after the last administration of dexamethasone. In addition, adequate cortisol suppression to the LDDST was assessed, using a cortisol value of less than 50 nmol/liter (<1.8 μg/dl) obtained at the same time, 48 h after the commencement of the LDDST (25). In 42 women with nontumorous hyperandrogenism who presented with an elevated testosterone, 21 of whom had previously failed to suppress testosterone during the LDDST, prednisolone (administered as 2.5 mg immediately on awakening and 5 mg immediately on retiring, using plain, nonenteric-coated tablets) was given for a minimum of 1 month. This dose was chosen to induce suppression of the normal nocturnal rise in ACTH, hence blocking activity of the pituitary-adrenal axis, while replacing glucocorticoid activity.

All hormonal assays were performed at the Department of Chemical Endocrinology at St. Bartholomew's Hospital (London, UK). Serum testosterone, androstenedione, and DHEAS were measured with in-house RIAs (24, 26).

Imaging

Ovarian ultrasonography. All five patients with ovarian androgen-secreting tumors had ovarian transabdominal ultrasonography (Table 1). These patients also underwent catheterization and sampling of the ovarian and adrenal veins to confirm the ovarian origin of excessive androgen production. In addition, in three patients with adrenal tumors, the transvaginal ultrasound appearance of the ovaries was suggestive of polycystic ovaries (PCO).

Among patients with nontumorous hyperandrogenism, 148 (70%) had either transabdominal or transvaginal ovarian ultrasonography. The diagnosis of PCO syndrome (PCOS) was based on the combination of clinical, endocrine, and ultrasonographic criteria (27). The presence of the ultrasonographic appearance of PCO (24) and the absence of any structural ovarian or adrenal abnormalities (PCOS may be copresent with androgen-secreting tumors) was taken to indicate nontumorous hyperandrogenism.

Adrenal computed tomography (CT) imaging. Similarly, all 12 patients with adrenal-androgen producing tumors had abnormal findings on adrenal CT scanning (Table 1). Among patients with nontumorous hyperandrogenism, 27 who failed to suppress their androgen levels on the LDDST had adrenal CT scanning to exclude adrenal pathology.

Histological features. Histological confirmation was obtained in all patients with tumorous hyperandrogenism. A further five patients with nontumorous hyperandrogenism had histological confirmation of PCOS: in two patients, surgery was performed to exclude the presence of an ovarian tumor, and in three as part of a therapeutic procedure (wedge resection).

Statistics

All data are presented as median values and range. The Kruskal-Wallis test was used for comparisons between independent groups. The Wilcoxon matched-pairs signed-rank sum test was used to compare within-subjects data before and after the LDDST. Correlation between variables was described using Spearman's rank correlation coefficients. Results were considered statistically significant if the P value was less than 0.05.

Results

Clinical features

The 211 women with nontumorous hyperandrogenism presented at a median age of 24 yr (range, 14–40 yr). The common presenting symptoms were hirsutism (95%), menstrual irregularity (65%), acne (29%), alopecia (10%), and clitoromegaly (8%). These patients fulfilled the clinical criteria for the diagnosis of PCOS (27, 28). In contrast, the five women with ovarian virilizing tumors presented at an older age (median, 45 yr; range, 18–62 yr; $P < 0.001$ compared with nontumorous women). Three of them presented with postmenopausal bleeding, but the other two had symptoms indistinguishable from patients with PCOS (Table 1). The 12 women with adrenal androgen-secreting tumors presented at a median age of 41 yr (range, 21–70 yr). Although two of them presented with symptoms suggestive of a virilizing tumor, the other three were initially diagnosed as having PCOS and were treated with antiandrogens for a median of 2 yr before referral to this institution.

Basal endocrine evaluation

The basal serum concentrations of each of the measured androgens in patients with tumorous and nontumorous hyperandrogenism overlapped (Table 2 and Fig. 1). Androstenedione was the most commonly elevated androgen in 96% of patients with nontumorous hyperandrogenism, followed by testosterone and DHEAS (47% in both; Table 2). Testosterone was the only androgen elevated in all patients with tumorous hyperandrogenism. Serum androstenedione was elevated in 11 of 14 (79%) and serum DHEAS in 11 of 16 (69%) patients with tumors (Table 1). Basal testosterone levels, although significantly higher in patients with tumors (median, 5.9 nmol/liter; range, 3.2–53.1 nmol/liter), overlapped with those obtained from patients with nontumorous hyperandrogenism (median, 2.8 nmol/liter; range, 1.0–7.3 nmol/liter; Fig. 1). Only testosterone levels greater than 7.3 nmol/liter distinguished patients with tumorous hyperan-

TABLE 1. Clinical and biochemical characteristics of the patients with androgen secreting tumors

Age	Symptoms	Pre-dexamethasone				Post-dexamethasone				Imaging	Histology
		T (nmol/liter)	A (nmol/liter)	DHEA (μmol/liter)	Cortisol (nmol/liter)	T (nmol/liter)	A (nmol/liter)	DHEAS (μmol/liter)	Cortisol (nmol/liter)		
29	H, A, C	13.2	183	19.9	296	9.7	156	14.9	162	CT: 11 cm mass L adrenal	Adrenal carcinoma
41	H, A, Al, OM, CS	6.8	32.4	11.3	699				674	CT: 6 cm mass R adrenal	Adrenal carcinoma
44	H, CS	3.9	20.0	17.4	1418				1595	CT: 12 cm R suprarenal mass	Adrenal carcinoma
27	H, A, C, OM	16		12.6						CT: 10 cm mass L adrenal	Adrenal carcinoma
42	H, CS	15.8	160	>27	1045					US: 10 cm R suprarenal mass	Adrenal carcinoma
34	H, A, AM, CS	3.2	42.2	>27	1201				917	US: 17 cm L suprarenal mass	Adrenal carcinoma
67	H, PMB, CS	22.2	>50	16.6	1245					CT: 11 cm mass L adrenal	Adrenal carcinoma
34	H, A, Al, AM	5.1	33	5.3	551	5.0	32	6.0	548	CT: 3.5 cm mass R adrenal	Adrenal carcinoma
61	H, Al	3.8	5.8	3.4		3.3				CT: 2 cm mass L adrenal	Adrenal carcinoma
70	H, HBP	53.1		49.2	548	52		47.2	713	CT: 4 cm mas R adrenal	Adrenal carcinoma
21	H, AM	3.6	15.5	4.6		2.9				CT: 2 cm mass L adrenal	Adrenal adenoma
43	H, C	4.5	35	23.5	385	4.2	35		<50	CT: 8 cm mass L adrenal	Adrenal adenoma
62	H, Al, PMB	4.8	16.0	1.4	730	5.9		0.7	<50	US: 2×2.5 bulky R ovary	Leydig cell tumor
26	H, C, OM	10.0	6.0	35.0	360	6.8	3.9	16	<50	US: 3.5×4 cm mass L ovary	Sertoli-Leydig cell tumor
59	H, C, HBP	13.3				15.0				US: very bulky ovaries	Hilus cell tumor bilateral stromal hyperplasia
18	H, A, Al, C	4.3	17.5	14.0	405	3.6	12.8	9.0	<50	US: R ovarian mass	Sertoli-Leydig cell tumor
61	H, PMB	13.0	7.0	3.4	270	10.0	6.5	3.3	<50	US: 5 cm mass L ovary	Granulosa cell tumor

Symptoms: H, hirsutism; A, acne; Al, alopecia; C, clitoromegaly; OM, oligomenorrhea; AM, amenorrhea; CS, Cushing's syndrome; HBP, high blood pressure; PMB, postmenopausal bleeding. Conversion factors: testosterone (nmol/liter; to convert to ng/ml, divide by 3.467); androstenedione (nmol/liter; to convert to ng/ml, divide by 3.492); DHEAS (μmol; to convert to μg/liter, divide by 368.46); cortisol (nmol/liter; to convert to μg/dl, divide by 27.59). US, Ultrasound; L, left; R, right.

TABLE 2. Circulating hormone levels in all women according to the etiology of hyperandrogenism

Variable	Nontumorous hyperandogenism (n = 211)	Ovarian tumors (n = 5)	Adrenal tumors (n = 12)
Testosterone (nmol/liter)	2.80 (1.0–7.3)	5.4 (3.5–13.3)	6.3 (3.2–53.1)
Androstendione (nmol/liter)	13.6 (4.0–35.0)	12.9 (6.0–17.0)	33.7 (5.8–183)
DHEAS (μmol/liter)	9.0 (1.4–38)	8.7 (1.4–35.0)	17.4 (3.4–49.2)
SHBG (nmol/liter)	26 (5–70)	33 (17–49)	36 (9–103)
17-OH-PG (nmol/liter)	5.3 (1.0–16.0)	7.0 (2.4–8.0)	21.8 (8.0–50)
Estradiol (pmol/liter)			
Premenopausal	201 (87–705)	130 and 337	175 (110–2650)
Postmenopausal		190 (140–290)	302 (85–1463)
LH (U/liter)			
Premenopausal	11.1 (2.0–49)	8.5 and 3.4	12.6 (0.3–26.1)
Postmenopausal		39 (18–50)	5.8 (0.3–49)
FSH (U/liter)			
Premenopausal	4.9 (0.9–10.7)	2.0 and 2.9	3.6 (0.4–18)
Postmenopausal		42 (15–50)	5.8 (0.2–50)
Prolactin (mU/liter)	273 (69–2914)	270 (142–603)	307 (166–446)

Data represent median (range). Conversion factors: testosterone (nmol/liter; to convert to ng/ml, divide by 3.467); androstenedione (nmol/liter; to convert to ng/ml, divide by 3.492); DHEAS (μmol; to convert to μg/liter, divide by 368.46); 17-OH-PG (nmol/liter; to convert to ng/ml, divide by 3.026); prolactin (mU/liter; to convert to ng/ml, divide by 20).

drogenism from patients with nontumorous hyperandrogenism, but such levels were encountered in only 41% of patients with tumors. Overall, a single elevated testosterone of over 3 nmol/liter was associated with a sensitivity of 100% in suggesting the presence of a virilizing tumor, but a specificity of only 53%. Factoring in the age variable and taking a cut-off of 40 yr, an age less than 40 yr and a testosterone greater than 3 nmol/liter had a 47% sensitivity and 53% specificity for an androgen-secreting tumor; an age above 40 yr and a similar testosterone greater than 3 nmol/liter had a sensitivity of 53% but a greatly increased specificity of 95%. Thus, a high testosterone in an older woman is of more sinister significance.

Of the 211 patients with nontumorous hyperandrogenism, 69% had at least two elevated androgens; testosterone plus androstenedione was elevated in 44%, whereas DHEAS plus androstenedione was elevated in 28% of patients, respectively. All three androgens were elevated in 20% of these patients. In contrast, 94% (15 of 16) of patients with tumorous hyperandrogenism had at least two elevated androgens, and 71% (10 of 14) had all three androgens elevated.

Analysis based on elevated basal serum androgen levels had a negative predictive value of 100% for serum testosterone, 97% for DHEAS, and 82% for androstenedione, with a positive predictive value of 15%, 12%, and 6%, respectively. Multiple linear logistic regression analysis including all three androgens showed that serum testosterone was the best predictor in distinguishing tumorous from nontumorous hyperandrogenism ($P < 0.05$). One of the two patients with basal serum 17-OH-PG greater than 7 nmol/liter, who subsequently underwent a short ACTH stimulation test, turned out to have late-onset congenital adrenal hyperplasia diagnosed on the basis of a peak 17-OH-PG level over 20 nmol/liter and was excluded from this study.

Androgen response during the 48-h LDDST

We analyzed the predictive value of the suppression of serum testosterone, because it was the only androgen consistently raised in all patients with tumors, during the LDDST obtained in patients with tumors compared with patients with nontumorous hyperandrogenism. As the greatest suppression of circulating testosterone, androstenedione, or DHEAS in patients with tumors was 32%, we chose a more severe threshold, a greater than 40% suppression or into the normal range, as indicating adequate androgen suppression. These responses were compared with those of patients with nontumorous hyperandrogenism.

Testosterone

All patients with tumors had elevated basal testosterone levels. No patient with a tumor achieved normalization of the previously elevated testosterone levels during the LDDST or a greater than 40% reduction from baseline. By contrast, 88% of women with nontumorous hyperandrogenism showed either a normalization of testosterone or suppression of more than 40% (Fig. 2). Thus, with respect to the above criterion of testosterone suppression, the LDDST was associated with 100% sensitivity and 88% specificity in identifying patients harboring androgen-secreting tumors.

DHEAS

Five patients with tumors had normal basal serum DHEAS levels (two with an ovarian tumor and three with an adrenal tumor) and a further two patients obtained adequate DHEAS suppression after the LDDST (two with an ovarian tumor). Two patients showed elevated levels of DHEAS that failed to suppress. Ninety-two percent of patients with nontumorous hyperandrogenism suppressed DHEAS adequately.

Androstenedione

Three patients with tumors had normal basal androstenedione levels, but none of the six patients with elevated androstenedione who underwent the LDDST obtained a 40% suppression or normalized the elevated basal androstenedione levels. However, 79% of women with nontumorous hyperandrogenism suppressed androstenedione appropriately (Table 3).

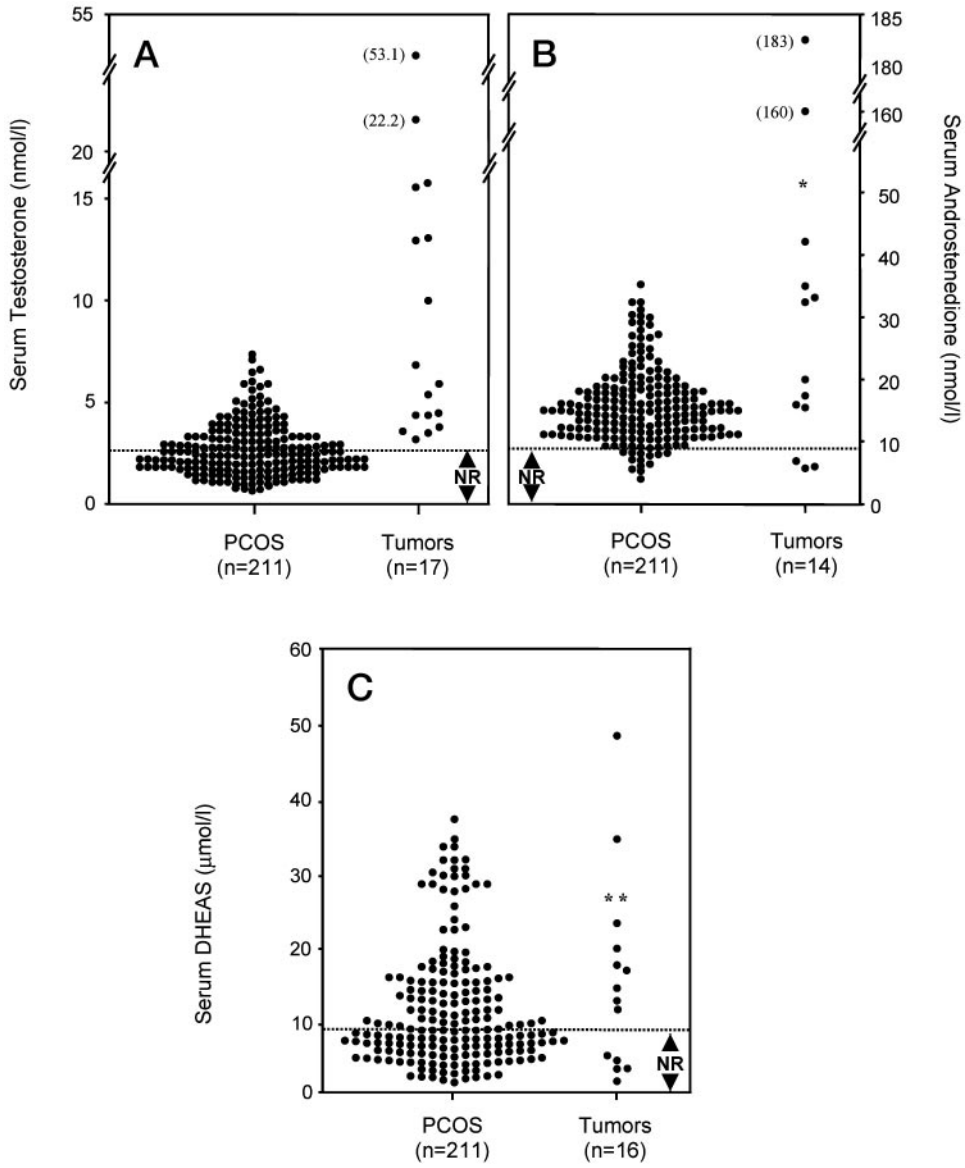

FIG. 1. Baseline serum testosterone (A), androstenedione (B), and DHEAS (C) levels in 228 women with hirsutism at presentation according to the cause of androgen excess. Conversion factors: testosterone (ng/ml = nmol/liter × 3.467^{-1}); androstenedione (ng/ml = nmol/liter × 3.492^{-1}); and DHEAS (μg/liter = μmol/liter × 368.46). NR, Normal range.

The combination of an elevated basal serum testosterone and a less than 40% suppression of serum testosterone during the LDDST offered the highest sensitivity and specificity for distinguishing tumorous from nontumorous hyperandrogenism. Because only 47% of women with nontumorous hyperandrogenism had elevated serum testosterone levels, we analyzed separately the subgroup of patients with nontumorous hyperandrogenism who had testosterone levels above 3 nmol/liter. The combination of an elevated basal serum testosterone and a greater than 40% reduction in circulating testosterone during the LDDST was associated with 100% sensitivity and 73% specificity in distinguishing virilizing tumors from other causes of hyperandrogenism.

One hundred fifty-two of 211 patients with nontumorous hyperandrogenism obtained a greater than 40% suppression or normalization in two or more of the measured androgens to the LDDST. Fifty-nine patients (28%) failed to obtain such a response in at least one androgen and 19 patients in more than one androgen, whereas only two patients failed to suppress all measured androgens (Table 4). Patients who failed to achieve adequate androgen suppression had significantly higher androgen levels, suggesting that androgen suppression to the LDDST may be related to baseline androgen levels. Although there was a weak but significant negative correlation between basal androgen levels and degree of suppression to the LDDST ($r = -0.16$; $P < 0.05$), there were individual patients with basal serum testosterone levels as high as 5.8 nmol/liter who achieved adequate suppression after dexamethasone administration.

The great majority, 95% (20 of 21), of patients with nontumorous hyperandrogenism who failed to achieve adequate testosterone suppression after the LDDST normalized all previously elevated androgens after treatment with prednisolone for 1 month (Fig. 3); no tumor was found in the 6-yr clinical and biochemical follow-up of the single patient who failed to normalize testosterone on prednisolone.

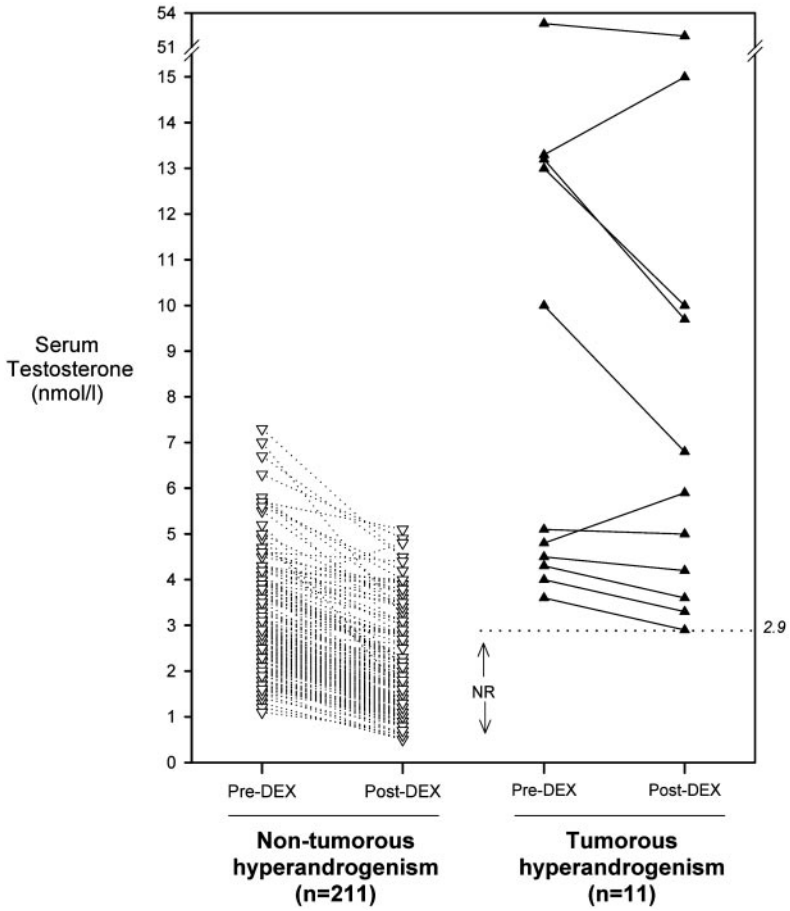

FIG. 2. Response of serum testosterone levels during the 48-h low-dose dexamethasone suppression test in 211 women with nontumorous hyperandrogenism and in 11 women with adrenal and ovarian androgen secreting tumors. *Solid line* in women with nontumorous hyperandrogenism is the median line. Conversion factors: testosterone (ng/ml = nmol/liter $\times$ 3.467^{-1}). NR, Normal range.

Cortisol suppression during the 48-h LDDST

Among patients with nontumorous hyperandrogenism, 209 of 211 (99%) demonstrated adequate cortisol suppression after the LDDST. However, two patients failed to obtain such a degree of suppression without any apparent reason for a false negative test, and no clinical evidence of Cushing's syndrome (25). Although both of these patients were lost to subsequent follow-up, neither had raised basal 0900 h serum cortisol levels (320 and 514 nmol/liter), nor a family history suggestive of a glucocorticoid-insensitivity syndrome.

Localization studies

Tumorous group. All five patients with ovarian androgen-secreting tumors had abnormal findings on ovarian ultrasonography (Table 1). Similarly, all 12 patients with adrenal-androgen producing tumors had abnormal findings on adrenal CT scanning (Table 1). In addition, in three patients with adrenal tumors, the transvaginal ultrasound appearance of the ovaries was suggestive of PCO.

Nontumorous group. Ovarian ultrasonography. One hundred and forty-eight (70%) patients had ovarian ultrasonography, and the diagnosis of PCOS was made in 125 (85%) of them. This included 50 of 59 patients who failed to obtain adequate androgen suppression after the LDDST who underwent ovarian ultrasonography, in which the presence of an ovarian androgen-producing tumor was excluded.

Adrenal CT imaging. Twenty-seven of the patients who failed to suppress their androgen levels on the LDDST had adrenal CT scanning; this was normal in 24 (89%). Two patients had findings consistent with mild nodular adrenal hyperplasia that remained stable in subsequent scans; these patients did not have evidence of autonomous cortisol secretion, in that there was complete suppression with dexamethasone administration. The other patient had an adrenal nodule, which enlarged on repeated scans but without evidence of endocrine hyperfunction; this patient underwent a unilateral adrenalectomy with histology consistent with an adrenocortical adenoma. All patients who failed to achieve adequate androgen suppression and did not have adrenal CT scanning normalized their elevated androgen levels after prolonged (>1 month) suppressive treatment with prednisolone.

Histological features. Histological confirmation was obtained in all patients with tumorous hyperandrogenism. Nine patients with adrenal tumors were found to have an adrenal carcinoma, and two patients had an adrenocortical adenoma. Five patients had a unilateral ovariectomy with histological confirmation of an ovarian tumor. Based on high clinical and biochemical suspicion and negative imaging and catheterization studies, two patients had laparoscopy and ovarian biopsy, whereas a further three patients underwent laparotomy with ovarian splitting and therapeutic wedge resection. All of these patients were shown to have PCOS on

TABLE 3. Hormone levels in serum of all women according to the etiology of hyperandrogenism, before (0 h) and after the administration of 2 mg/d dexamethasone for 48 h

Variable	Nontumorous hyperandrogenism (n = 211) Median (range)	Tumorous hyperandrogenism (n = 11) Median (range)
Testosterone (nmol/liter)		
0 h	2.60 (1.1–7.3)	5.1 (3.6–53.1)
48 h	1.40 (0.5–5.1)	5.9 (2.9–52.0)
% change	−43 (−54 to +16)	−17 (−32 to +23)
Androstendione (nmol/liter)		
0 h	13.1 (4.1–34.0)	17.5 (5.8–183)
48 h	6.20 (0.9–31.0)	22.4 (3.9–156)
% change	−52 (−90 to +20)	−11 (−35 to 0)
DHEAS (μmol/liter)		
0 h	9.30 (1.7–37.0)	16.9 (1.4–49.2)
48 h	3.30 (0.5–31.0)	9.0 (0.7–47.2)
% change	−61 (−97 to +23)	25 (−54 to +13)
Cortisol (nmol/liter)		
0 h		
NTH	396 (110–906)	
Ovarian T		382 (247–730)
Adrenal T		551 (296–980)
48 h		
NTH	<50	
Ovarian T		<50
Adrenal T		445 (78–713)

Data represent median (range). T, Tumor; NTH, nontumorous hyperandrogenism. Conversion factors: testosterone (nmol/liter; to convert to ng/ml, divide by 3.467); androstenedione (nmol/liter; to convert to ng/ml, divide by 3.492); DHEAS (μmol; to convert to μg/liter, multiply by 368.46); cortisol (nmol/liter; to convert to μg/dl, divide by 27.59).

TABLE 4. Basal and dexamethasone-suppressed hormone levels in blood of all women with nontumorous hyperandrogenism differentiated according to the response of serum androgen levels to administration of dexamethasone (2 mg/d for 48h)

	Women who suppressed all measured androgens (n = 152)	Women who failed to suppress at least one of the measured androgens (n = 59)
Baseline hormone levels		
LH (U/liter)	9.3 (2.5–49)	13.4 (2.0–37.5)[b]
FSH (U/liter)	4.6 (0.9–10.7)	5.0 (1.7–8.4)
Testosterone (nmol/liter)	2.5 (1.0–5.8)	3.3 (1.8–7.3)[b]
Androstendione (nmol/liter)	13.5 (4.0–29.2)	14.2 (5.6–35.0)[a]
DHEAS (μmol/liter)	9.0 (1.7–38.0)	9.3 (1.4–35.0)
Estradiol (pmol/liter)	197 (87–705)	216 (99–574)
SHBG (nmol/liter)	26 (5–70)	25.5 (10–52)
Dexamethasone suppression test		
Testosterone 0 h	2.4 (1.1–5.8)	3.5 (2.2–7.3)[b]
Testosterone 48 h	1.3 (0.5–3.3)	2.8 (1.0–5.1)[b]
Androstendione 0 h	12.6 (4.1–31.2)	14.0 (5.4–34.0)[a]
Androstendione 48 h	5.4 (0.9–18.8)	9.8 (1.5–31.0)[b]
DHEAS 0 h	9.2 (1.7–37)	9.4 (2.7–31.0)
DHEAS 48 h	3.2 (0.5–18.0)	4.0 (0.5–31.0)[a]
Cortisol 0 h (nmol/liter)	414 (110–906)	370 (115–870)[a]
Cortsol 48 h	<50 (all but 3)	<50 (all but 2)

Data represent median (range). Mann-Whitney test: suppression *vs.* failure to suppress; [a] $P < 0.05$; [b] $P < 0.01$.

histology, and no androgen-secreting tumor embedded within the ovary was found.

Discussion

The presence of serum testosterone concentrations well above the normal range in women presenting with hyperandrogenism is often regarded as a particularly reliable indicator of the presence of a virilizing tumor of either the ovaries or the adrenal cortex (2, 3, 5). Although all patients with androgen-producing tumors in this relatively small series had elevated basal testosterone levels, only testosterone values greater than 7.3 nmol/liter fully discriminated such patients from those with nontumorous hyperandrogenism; however, over half of the patients with virilizing tumors had only marginally elevated testosterone levels well within the range observed in patients with nontumorous hyperandrogenism. This emphasizes the limitation of basal serum testosterone measurement alone in identifying patients harboring virilizing tumors. Furthermore, only mildly elevated or even normal DHEAS levels were found in this series of patients, and measurement of basal DHEAS levels was the least effective in identifying patients with tumors (29). How-

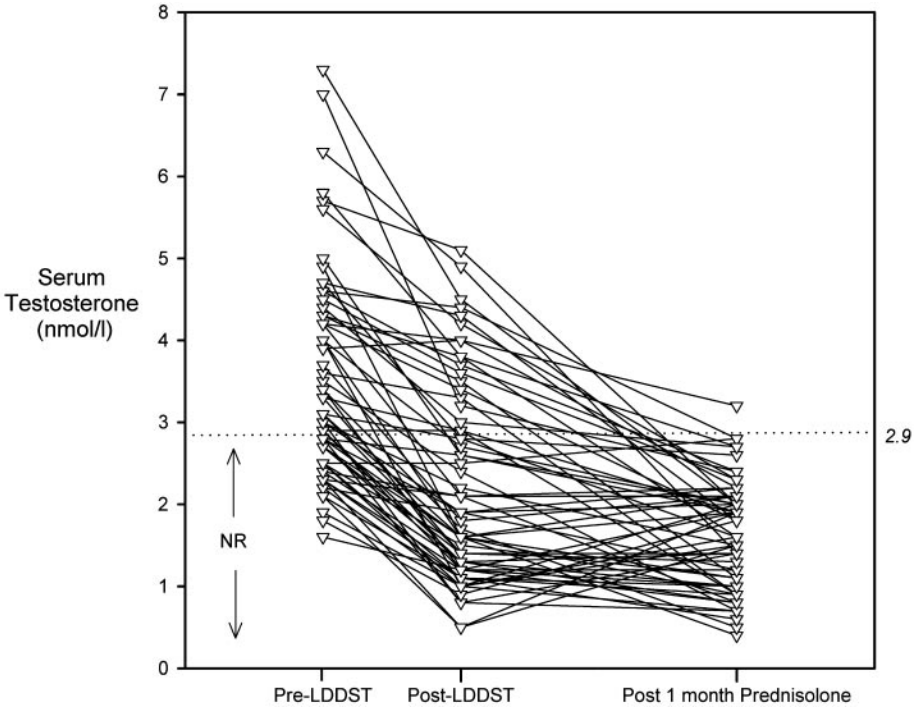

FIG. 3. Response of serum testosterone levels during the 48-h low-dose dexamethasone suppression test and after a 4-wk treatment with reverse circadian rhythm prednisolone in 42 women with nontumorous hyperandrogenism. Conversion factors: testosterone (ng/ml = nmol/liter $\times$ 3.467^{-1}). NR, normal range.

ever, because tumors preferentially secreting androgens other than testosterone have been described elsewhere (7), it could be argued that measurement of all three androgens is still indicated. Measurement of 17-OH-PG is also valuable in identifying the nontumorous patients with late-onset congenital adrenal hyperplasia (2).

It has been suggested that virilizing tumors can be detected on purely clinical grounds without extensive hormonal testing (4). A short history of amenorrhea and rapid progression of hirsutism and virilization (male-pattern baldness, muscle development, deepening of the voice) has been thought to be sufficient to identify virilizing tumors, especially when this progression begins outside the peripubertal years (3, 5). However, five of the patients in this series with androgen-secreting tumors had a gradual onset of symptoms and a clinical presentation identical to patients with nontumorous hyperandrogenism, and were originally thought elsewhere to have PCOS. It was only when there was worsening of their clinical and hormonal features, despite treatment, that the correct diagnosis was made. The latter may be due to the fact that 3 of 12 (25%) of the patients in this series with adrenal carcinoma had an ovarian ultrasound appearance suggestive of PCOS, which delayed further investigations (26). In addition, the data provided in this series show that patients with tumors, despite elevated androgen levels, may sometimes present with elevated gonadotropin levels and an increased LH-to-FSH ratio that can be interpreted as suggestive of PCOS. Therefore, reliance on clinical presentation and basal androgen measurements alone is insufficient to identify or reliably exclude the presence of virilizing tumors in patients presenting with hirsutism.

After the 48-h LDDST, none of the patients with androgen-secreting tumors normalized their previously elevated serum testosterone levels or reduced these levels by 40% or more; only 12% of patients with nontumorous hyperandrogenism failed to achieve either one or the other of such responses. Thus, in the current series the 48-h LDDST offers 100% sensitivity in identifying patients with androgen-secreting tumors with a relatively high specificity (88%). Although this number is considerably lower than the 100% sensitivity and specificity obtained in a previous study using more prolonged dexamethasone administration (3 mg/d for 5 d), that study included only patients with adrenal androgen-producing tumors (6). The finding of the current study is particularly important because most adrenal androgen-producing tumors are relatively large at presentation and can be readily detected by newer imaging modalities (30–32). By contrast, ovarian tumors may be relatively small, occasionally embedded within ovaries with a polycystic appearance, and may elude detection with the common imaging modalities, although the application of newer more sensitive techniques, such as high-resolution transvaginal ultrasound with Doppler studies, was not routinely performed in this series (30–32). In addition, 95% of the patients with nontumorous hyperandrogenism who failed to achieve adequate androgen suppression after the 48-h LDDST and received prolonged treatment with glucocorticoids normalized their elevated androgens (8, 9, 33). This treatment protocol was designed to suppress the normal pituitary-adrenal axis and thus allow the adrenal contribution to the PCOS to be nullified. Although we did not specifically treat the androgen-secreting tumors in a similar manner in our series, no patient with an androgen-secreting tumor has been reported to achieve a similar response. We therefore suggest that the addition of one month's treatment with an adrenal-suppressing corticosteroid regimen may improve the specificity for serum androgens in those patients who did not show suppression during the LDDST and had negative imaging stud-

ies of the ovarian and adrenal glands, but in whom the clinical/biochemical suspicion of an androgen-producing tumor remains high.

Although there was a weak negative correlation between basal androgen levels and the degree of androgen suppression to the 48-h LDDST, basal androgen levels in general cannot be used to predict the response to dexamethasone suppression. Lack of androgen suppression to the 48-h LDDST strongly raises the possibility of an androgen-secreting tumor of either the ovaries or the adrenal glands; however, ovarian and adrenal venous catheterization and sampling did not reveal the presence of a virilizing tumor in any of these patients when ovarian or adrenal imaging was negative (our manuscript in preparation). We therefore suggest that further investigations should be undertaken only in patients with hyperandrogenism who fail to achieve 40% or more testosterone suppression during a 48-h 2 mg/d LDDST, and this should initially be limited to ovarian and adrenal imaging. Additional investigation should be undertaken only when androgen levels fail to suppress to within the normal range after more prolonged glucocorticoid suppressive treatment (8, 9). Most of the patients presenting with hyperandrogenism will be found to have PCOS as the underlying diagnosis (28, 34). This is once again demonstrated in this series of 211 patients in which the clinical, endocrine, radiological, and histological features were consistent with the syndrome (27, 28). The findings of this series suggest that the 48-h LDDST is a valuable simple tool in the investigation of hyperandrogenic women, because it can also identify with 98% sensitivity women with Cushing's syndrome and patients with alterations of glucocorticoid distribution or metabolism by means of adequate cortisol suppression (25).

Hyperandrogenism has been linked to excessive androgen production from the ovary or the adrenal, or from peripheral conversion from precursors (2, 3). Although there is considerable evidence implicating the adrenal glands as a source of excessive androgen production in such patients, studies looking at steroid output after catheterizing such patients and androgen production rates implicated the ovary as the major source, although there is often an additional significant adrenal component (1, 9, 33). Recently, a unifying hypothesis implicating both the adrenal and the ovary in the pathogenesis of nontumorous hyperandrogenism has been described; it has been postulated that dysregulation of androgen formation by 17-hydroxylase and 17,20-lyase, prominently involving cytochrome P450c17, affects both adrenal and ovarian steroidogenesis (35). Thus, it is postulated that the ability of glucocorticoids to affect both androgen-producing tissues can be used to differentiate between the causes of hyperandrogenism.

In summary, although clinical presentation alone may be highly suggestive of a virilizing tumor, it can also be misleading, particularly in premenopausal hyperandrogenic women. Although elevated serum testosterone levels were found in all patients in this relatively small series of patients with virilizing tumors, this may only be marginally above normal. Serum basal testosterone levels that are elevated but less than 7.3 nmol/liter occur in both patients with androgen-secreting tumors and patients with nontumorous hyperandrogenism. In women with clinical features of hyperandrogenism, a greater than 40% suppression or normalization of serum testosterone after the LDDST was associated with 100% sensitivity and 88% specificity in distinguishing patients with ovarian and adrenal androgen-secreting tumors from patients with nontumorous hyperandrogenism. A 4-wk treatment with prednisolone 2.5 mg on awakening and 5 mg on retirement is associated with suppression of circulating androgens in over 95% of patients with nontumorous hyperandrogenism who have high clinical and/or biochemical suspicion of an androgen-producing tumor but negative imaging studies. In addition, adequate cortisol suppression to the LDDST can detect patients with Cushing's syndrome and also identify patients with glucocorticoid insensitivity or alterations of glucocorticoid metabolism.

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Address all correspondence and requests for reprints to: Prof. Ashley B. Grossman, Endocrine Oncology, Department of Endocrinology, St. Bartholomew's Hospital, London EC1A 7BE, United Kingdom. E-mail: A.B.Grossman@qmul.ac.uk.

Present address for A.M.I.: Cattedra di Andrologia, Dipartimento di Fisiopatologia Medica, Università "La Sapienza" di Roma, 2 00185 Roma, Italy.

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