

A Single Midnight Serum Cortisol Measurement Distinguishes Cushing's Syndrome from Pseudo-Cushing States*

DIMITRIS A. PAPANICOLAOU, JACK A. YANOVSKI, GORDON B. CUTLER, JR.†, GEORGE P. CHROUSOS, AND LYNNETTE K. NIEMAN

Developmental Endocrinology Branch, National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, Maryland 20892

ABSTRACT

Cushing's syndrome (CS) may be difficult to distinguish from pseudo-Cushing states (PCS) based on physical findings or urinary glucocorticoid excretion. As the lack of diurnal variation in serum cortisol is characteristic of CS, we studied whether diurnal cortisol determinations could discriminate CS from PCS. Two hundred and sixty-three patients were evaluated: 240 had CS, and 23 had PCS. Urine was collected for 24 h for measurement of cortisol and 17-hydroxycorticosteroids (17OHCS). Blood was drawn at 2300, 2330, 0000, 0030, and 0100 h and at 0600, 0630, 0700, 0730, and 0800 h the next morning for serum cortisol determination. The main outcome measure was the sensitivity of these parameters for the diagnosis of CS

at 100% specificity. A midnight cortisol value greater than 7.5 $\mu\text{g}/\text{dL}$ correctly identified 225 of 234 patients with CS and all PCS patients. This sensitivity (96%) was superior to that obtained for any other measure, including urinary cortisol (45%), 17OHCS (22%), any other individual cortisol time point (10–92%), the morning (23%) or the evening (93%) cortisol mean, and the ratio (11%) of morning to evening values. We conclude that at 100% specificity, a single serum cortisol value above 7.5 $\mu\text{g}/\text{dL}$ at midnight discriminates CS from PCS with higher sensitivity than 24-h urinary cortisol or 17OHCS, or other individual or combined measures of serum cortisol. (*J Clin Endocrinol Metab* 83: 1163–1167, 1998)

OVERPRODUCTION of cortisol is the biochemical hallmark of Cushing's syndrome (CS) regardless of its etiology and is evidenced by increased urinary cortisol excretion, and a decrease in the circadian variation of serum cortisol (1).

Pseudo-Cushing states (PCS), as the name implies, share many of the features of Cushing's syndrome, including cortisol overproduction. The hypercortisolism of PCS is hypothesized to be caused by increased activity of the CRH neuron, which, in turn, stimulates ACTH production and release (2). PCS are a heterogeneous group of disorders, including chronic alcoholism and alcohol withdrawal syndrome (3, 4), major depression (5), poorly controlled diabetes mellitus (6, 7), and obesity (8). Additionally, transient hypercortisolism may be associated with less obvious psychiatric conditions (e.g. anxiety) in patients with clinical features reminiscent of CS, such as obesity and hypertension, which are common in the general population. The substantial overlap in urinary free cortisol (UFC) excretion and clinical features between some patients with CS and those with PCS can make it difficult to distinguish between the two conditions (9). Thus,

although persistent elevation of 24-h UFC in the presence of unequivocal signs of CS (particularly classic moon facies, prominent centripetal obesity, severe proximal muscle weakness, and violaceous striae) suggest the diagnosis of CS, patients with less obvious signs pose a diagnostic dilemma.

Several tests have been proposed to diagnose CS, including 24-h UFC measurements, the 1-mg overnight dexamethasone suppression test (DST) (10), the 2-day DST (1), and the dexamethasone-CRH (Dex-CRH) stimulation test (8). Each has drawbacks. Twenty-four-hour urinary collections are inconvenient and often incomplete. The 1-mg overnight DST is commonly used as a screening test to exclude the diagnosis of CS. This test has two caveats. First, a criterion for the level of serum cortisol suppression to exclude CS has not been developed using modern RIAs. Second, although the test has a false negative rate of only 2%, it has a significant false positive rate, especially in chronically ill (23%) or obese patients (13%) (11) and in patients with major depression (43%) or other psychiatric disorders (8–41%) (12). Even in normal individuals, the test may be consistent with CS in up to 30% (9).

Similarly, the 2-mg 2-day DST, often used as a confirmatory diagnostic test, has a diagnostic accuracy of only 71% (8). An additional problem is the variable metabolic clearance of dexamethasone (13), which is especially problematic in patients receiving medications that induce the cytochrome P450-related enzymes (e.g. phenytoin, rifampin, and phenobarbital) (14) or in patients with renal or hepatic failure. In such cases, neither DST gives reliable results. Finally, the drawbacks of 24-h urine collections apply to the DST as well.

We previously determined that the dexamethasone-CRH

Received October 22, 1997. Revision received December 2, 1997. Accepted January 5, 1998.

Address all correspondence and requests for reprints to: Dimitris A. Papanicolaou, M.D., Developmental Endocrinology Branch, National Institute of Child Health and Human Development, National Institutes of Health, Building 10, Room 10N262, 10 Center Drive, MSC 1862, Bethesda, Maryland 20892-1862. E-mail: papanicd@cc1.nichd.nih.gov.

* Presented at the 76th Annual Meeting of The Endocrine Society, Anaheim, CA, 1994.

† Current address: Eli Lilly Co., Lilly Research Laboratories, Indianapolis, Indianapolis 46285-2055.

test has a diagnostic accuracy of 98% in the distinction of CS from PCS (8, 15). However, although accurate, this test has the drawbacks related to dexamethasone clearance, as discussed above.

Physiological cortisol secretion is characterized by circadian rhythmicity. Serum cortisol concentration reaches its zenith in the morning (0600–0800 h) and its nadir in the night during the first half of normal sleep. Krieger *et al.* defined the normal circadian rhythm of plasma corticosteroid levels as the pattern where all plasma glucocorticoid levels from 1600–2400 h were 75% or less of the 0800 h value (16). As previous studies have found that obese individuals retain a normal circadian cortisol rhythm (17), we hypothesized that differences in circadian plasma cortisol values would distinguish CS from PCS. To test this hypothesis, we prospectively measured serum cortisol values during the normal nadir and zenith periods in patients being evaluated for CS.

Subjects and Methods

Subjects

Three hundred and twenty-two consecutive patients admitted for the evaluation of CS to the Clinical Center of the NIH between June 1987 and April 1992 participated in the study, after giving informed consent. Patients had been accepted for evaluation based on clinical findings suggestive of CS and either elevated 24-h UFC or failure to suppress morning serum cortisol after oral administration of 1 mg dexamethasone at 2300–2400 h the previous evening. After initial evaluation, patients were followed until a diagnosis of CS or PCS was made. The diagnosis of CS was based on tissue pathology or hypocortisolism after surgery (transsphenoidal surgery, adrenalectomy, or removal of tumor producing ectopic ACTH) or on resolution of clinical and laboratory abnormalities after medical, surgical, or radiation therapy. The diagnosis of PCS was based on negative tests for CS (including a dexamethasone-CRH stimulation test, as previously described) (8) and no progression of clinical or biochemical characteristics on follow-up of at least 1 yr (mean, 21 months; range, 12–42 months).

Based on the above diagnostic criteria, 59 of 322 patients were excluded from analysis; 11 had undergone radiation therapy to the pituitary gland before admission, 7 were receiving medical therapy for CS at the time of evaluation, 1 had surreptitious ingestion of cortisol, and in 40 the diagnosis was not established [12 patients, referred for possible recurrence of CS after transsphenoidal surgery (TSS) for Cushing's disease, either received no treatment at the NIH or had inadequate follow-up; 3 patients had incomplete studies; 6 had no treatment at the NIH for presumed CS; in 6 no tumor was found during TSS, and follow-up was inadequate; in 13 patients the diagnosis of CS *vs.* PCS could not be made due to inadequate length of follow-up].

Protocol

Urine was collected for 24 h, starting at 0600 h, for measurement of UFC, 17-hydroxycorticosteroids (17OHCS), and creatinine. On the same day, an indwelling venous catheter was placed at 2200 h, and blood was drawn for serum cortisol determination at 2300, 2330, 0000, 0030, and 0100 h and at 0600, 0630, 0700, 0730, and 0800 h the next morning. As food ingestion may be associated with elevation of the circulating cortisol concentration (18), all patients were fasting from 2100 h until the conclusion of sampling. Patients traveling from a time zone different

than that of Bethesda, MD were studied at least 3 days after arrival. One of the 23 patients with PCS and 19 of the 240 patients with CS did not have UFC measurements on the same day as the serum cortisol determinations. The patient with PCS had elevated 17OHCS (28.2 mg/day; 12.4 mg/g creatinine), and of the 19 patients with CS, 7 had elevated 17OHCS on the day serum cortisol was measured. Twelve patients with CS did not have urine collected during the serum cortisol measurements. The midnight serum cortisol measurement was not available for 6 of the 240 patients with CS.

Assays

Serum cortisol was measured by RIA (Premix kit, Diagnostic Products Corp., Los Angeles, CA) until January 1988, and by fluorescence polarization immunoassay (Abbott Laboratories, Abbott Park, IL) afterward. The intra- and interassay coefficients of variation were 3.8% and 5.8% for the RIA and 2.1% and 4.1% for the fluorescence polarization immunoassay, respectively. The two assays had similar performance characteristics. UFC was measured by RIA (SmithKline Bioscience Laboratories, King of Prussia, PA). The intra- and interassay coefficients of variation were 5.4% and 9.3–11.5%, respectively. Urinary 17OHCS was measured using the Porter-Silber method (intra- and interassay coefficients of variation, 5.9% and 10.7%, respectively).

Statistical analysis

The following measures were evaluated: serum cortisol at 10 individual time points, the mean serum cortisol of the 5 morning time points (AM), the mean serum cortisol of the 5 evening time points (PM), the difference (AM-PM) and the ratio between these means (AM/PM), the 24-h UFC, and 24-h urinary 17OHCS excretion per g 24-h urinary creatinine. Values are expressed as the mean \pm SEM.

Data were analyzed using repeated measures ANOVA (with logarithmic transformation where appropriate), followed by Student-Newman-Keuls *post-hoc* test. We used the cut-off criterion that gave 100% specificity for the diagnosis of CS to calculate the sensitivity of each parameter for the diagnosis of CS. We assessed the diagnostic value of each parameter in the entire group as well as in the subset of patients with mild to moderate hypercortisolism, which included all patients with PCS and only those patients with CS whose UFC was within the range seen in patients with PCS (*i.e.* up to 389 μ g/day). Receiver operator characteristic (ROC) curves were used to assess the utility of each measure for the distinction of CS from PCS (19).

Results

Patients

The clinical characteristics of the 263 patients suitable for analysis are shown in Table 1. Two hundred and forty patients had CS (Cushing's disease, $n = 198$; ectopic ACTH production, $n = 27$; primary adrenal disease, $n = 15$), and 23 had PCS. There was a female predominance in each diagnostic category. No age difference was noted between the CS and the PCS groups ($P = 0.56$ and 0.26 for females and males, respectively). Approximately two thirds of all patients (68% of PCS and 69% of CS) were obese (body mass index, >27 kg/m²). Adult patients with PCS weighed more than adult patients with CS (82.9 ± 1.6 kg for CS *vs.* 99.1 ± 6.7 kg for PCS; $P = 0.04$ for males and females), but the difference in

TABLE 1. Patient characteristics

	Age yr (range)	Gender, M/F (%F)	Wt (kg)	Ht (m)	BMI (kg/m ²)
CS	35 (5–77)	59/181 (75)	82.9 \pm 1.6 ^a	1.62 \pm 0.01	31.4 \pm 0.5
PCS	34 (17–61)	8/15 (65)	99.1 \pm 6.7	1.67 \pm 0.02	35.6 \pm 2.3

CS, Cumulative data for CS of any etiology.

^a $P < 0.05$ *vs.* PCS.

BMI did not reach statistical significance ($P = 0.05$ and 0.11 for females and males, respectively).

Additional diagnoses were present in 20 of 23 patients with PCS. Sixteen patients had psychiatric evaluation either at the Clinical Center or by the referring physicians: 7 were diagnosed with depression, 4 were diagnosed with depression combined with other psychiatric diseases (1 with anorexia nervosa and panic disorder, 1 with somatoform disorder, 1 with obsessive compulsive disorder, and 1 with drug dependence), 1 was diagnosed with bipolar disorder and alcoholism, 1 with avoidant personality disorder, 1 with adjustment disorder, 1 with somatization disorder, and 1 with dysthymic disorder. One patient had poorly controlled diabetes mellitus (hemoglobin A_{1c}, 12.2%). Three patients presented with elevated UFC 2, 3, and 4 yr, respectively, after successful transsphenoidal surgery for Cushing's disease. Recurrent Cushing's disease was excluded after detailed and repeated evaluation for 3 additional yr, showing no progression of clinical features, repeatedly normal UFC excretion, and in one case a normal response to a dexamethasone-CRH stimulation test. Finally, in 3 patients no additional medical problem was identified besides obesity and physical stigmata, warranting the evaluation of possible CS.

Biochemical data

Twenty-four-hour excretion of glucocorticoids was as follows: UFC, 695 ± 84 (range, 12–9703) and 135 ± 18 (range, 48–388) $\mu\text{g}/\text{day}$ for CS and PCS patients, respectively ($P < 0.00001$); 17OHCS, 24.8 ± 1.7 (range, 2.3–265) and 12.0 ± 1.2 (range, 4.5–28) mg/day for CS and PCS patients, respectively ($P < 0.00001$); and 17OHCS per g urinary creatinine, 21.5 ± 2.1 (range, 1.9–319) and 7.9 ± 0.6 (range, 3.8–16) for CS and PCS patients, respectively ($P < 0.00001$). The highest UFC measured in a patient with PCS was 388 $\mu\text{g}/\text{day}$. The serum cortisol measurements for the 10 time points are shown in Fig. 1. The ratios between the average morning and evening serum cortisol concentrations were 1.18 ± 0.06 (range, 0.4–12.2) and 5.38 ± 0.61 (range, 0.9–12.7) for CS and PCS, respectively ($P < 0.00001$). The Δ values (average morning

minus average evening serum cortisol value) were 1.90 ± 0.35 (range, –14.9 to 29.8) and 10.81 ± 1.13 (range, –1.3 to 21.7) for CS and PCS, respectively ($P < 0.00001$).

Criteria for the diagnosis of CS

The sensitivity for the diagnosis of CS, at 100% specificity for each of the diagnostic parameters, is shown in Table 2. Of all parameters studied, a midnight serum cortisol concentration of more than 7.5 $\mu\text{g}/\text{dL}$ (19.8 ± 0.9 (range 1.3–122.6) and 3.1 ± 0.3 (range, 1–7.5) for CS and PCS, respectively) gave the highest sensitivity [96%; confidence interval (CI), 94–99%] for the diagnosis of CS, at a specificity of 100%. The diagnostic value of the single midnight serum cortisol concentration remained virtually unchanged (95%; CI, 91–99%) when only patients with 24-h UFC less than 389 $\mu\text{g}/\text{day}$ were analyzed. However, the sensitivity of the midnight serum cortisol concentration was only 89% (CI, 81–97%) when only patients with mild hypercortisolism (*i.e.* UFC, $<200 \mu\text{g}/\text{day}$) were analyzed. The scatterplot of the midnight serum cortisol concentration for patients with CS and PCS is shown in Fig. 2, and the ROC curve for the same parameter in Fig. 3.

Twenty-two patients with CS had UFC within the normal range ($\leq 90 \mu\text{g}/\text{day}$) at the time of study. A midnight serum cortisol measurement correctly identified 20 of these patients as well as 17 PCS patients with elevated UFC at the time of study (Fig. 4). None of the other parameters studied, including UFC, 17OHCS, AM/PM ratio, AM-PM Δ , or individual time points, reached that level of diagnostic accuracy.

Discussion

In this study, a midnight serum cortisol concentration of more than 7.5 $\mu\text{g}/\text{dL}$ identified correctly 96% of 234 patients with CS. In comparison, UFC, 17OHCS, and 17OHCS per g excreted creatinine were less helpful as diagnostic tools for the differential diagnosis of CS *vs.* PCS. In fact, the majority

TABLE 2. Diagnostic value of biochemical parameters for the diagnosis of CS, using a criterion that achieved 100% specificity

Parameter	Criterion	Sensitivity (%)	
		All patients (n = 263)	Patients with UFC $\leq 388 \mu\text{g}/\text{day}$ (n = 144)
UFC ($\mu\text{g}/\text{day}$)	>388	45	0
17OHCS (mg/day)	>28.2	22	3
17OHCS/Cr ($\mu\text{g}/\text{day} \cdot \text{g}$)	>16.0	47	27
F 0600 h	>31.1	10	3
F 0630 h	>27.4	16	6
F 0700 h	>23.5	28	13
F 0730 h	>24.9	21	9
F 0800 h	>26.5	19	9
F 2300 h	>14.5	73	60
F 2330 h	>9.1	92	87
F 0000 h	>7.5	96	95
F 0030 h	>11.9	85	78
F 0100 h	>20.6	34	18
AM	>24.22	23	11
PM	>9.96	93	89
AM/PM	<0.87	11	16
AM-PM	<-1.28	22	24

17OHCS/Cr, 17OHCS per g urinary creatinine (milligrams per day/g); F, serum cortisol (micrograms per dL); AM and PM, average morning and evening serum cortisol concentrations, respectively.

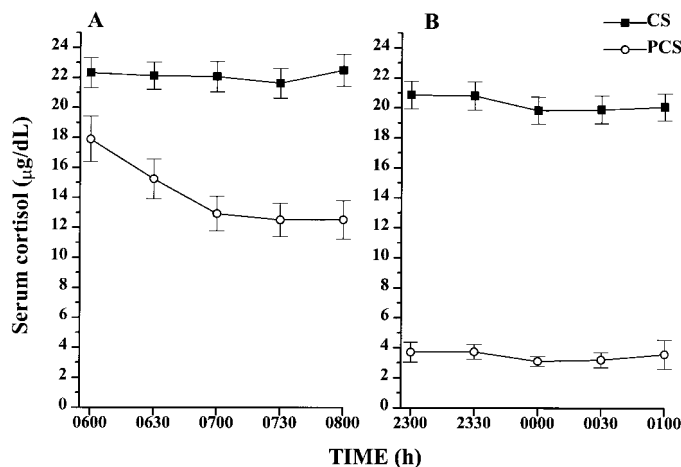


FIG. 1. Morning (A) and evening (B) serum cortisol concentrations in patients with CS (solid squares) and in patients with PCS (open circles). $P < 0.00005$ (CS *vs.* PCS) for all time points, by repeated measures ANOVA, followed by Student-Newman-Keuls test.

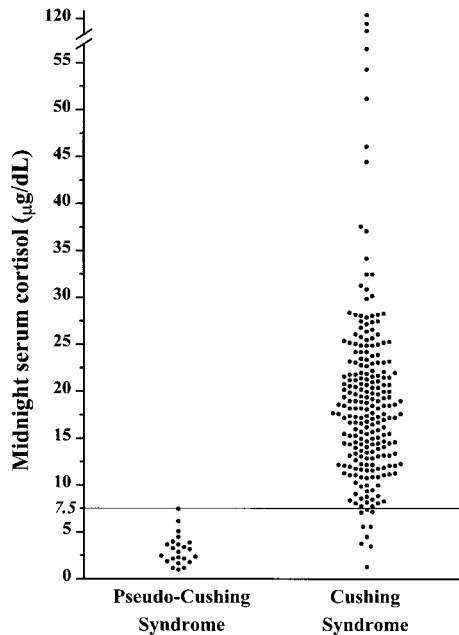


FIG. 2. Midnight (0000 h) serum cortisol concentration in patients with CS and PCS. The line represents the criterion value of 7.5 $\mu\text{g/dL}$ for the diagnosis of CS.

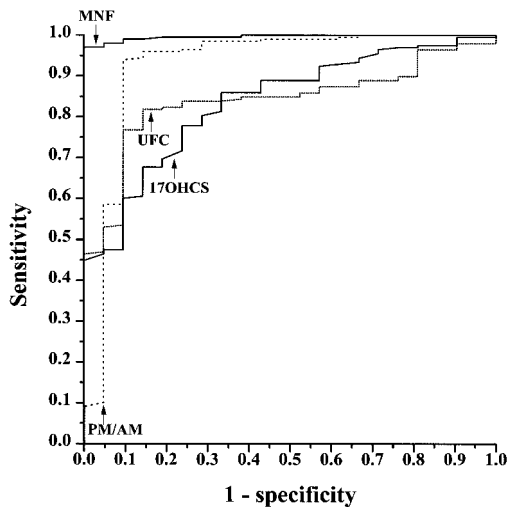


FIG. 3. ROC curve using midnight serum cortisol concentration (MNF), UFC, 17OHCS and PM/AM ratio as criteria for the diagnosis of CS.

of patients with CS (55%) had UFC levels within the range seen in patients with PCS, *i.e.* less than 389 $\mu\text{g/day}$. In contrast to the discrimination by midnight serum cortisol, morning serum cortisol did not distinguish between these entities, consistent with previous reports (20). Interestingly, neither the ratio nor the Δ between the average morning and the average nighttime serum cortisol values was helpful in the diagnosis of CS. It has been proposed that any evening (after 1600 h) serum cortisol concentration greater than 75% of the 0800 h concentration is highly suggestive of CS (16). When this criterion was applied to our cohort, the test sensitivity and specificity for the diagnosis of CS were 71–76% and 83–96%, respectively. Our data confirm previous observa-

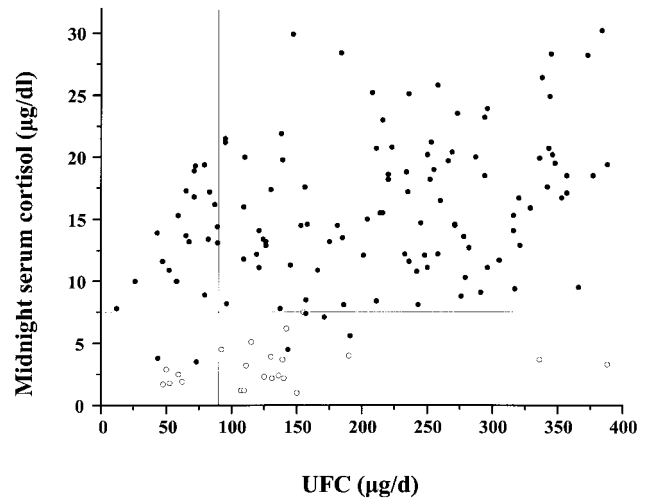


FIG. 4. UFC vs. midnight serum cortisol in patients with UFC below 400 $\mu\text{g/day}$. The vertical line represents the upper limit of normal for UFC (90 $\mu\text{g/day}$), and the horizontal line represents the cut-off value of 7.5 $\mu\text{g/dL}$ for midnight serum cortisol. Solid circles, CS; open circles, PCS.

tions that the circadian rhythm of cortisol secretion is retained in a subgroup of patients with CS, but appears to be set at a higher point (17, 21, 22) or is phase shifted (23). Thus, this study supports the idea that the most consistent circadian abnormality in CS is a blunted midnight nadir.

In this study, we inserted the indwelling iv catheter for drawing blood at least 1 h before sampling, as the stress of the direct venipuncture might be sufficient to raise serum cortisol values. Patients were fasting from 2100 h until the conclusion of sampling, because food ingestion could also increase serum cortisol levels. All patients were instructed to avoid any physical activities for at least 2 h before sampling. Therefore, our conclusions with regard to midnight serum cortisol levels in CS and PCS apply only when there are similar restrictions on diet and activity.

Recently, based on plasma cortisol levels in normal volunteers of 2 $\mu\text{g/dL}$ at midnight, this value has been suggested as the cut-off value for the diagnosis of CS, as all 150 patients with CS examined had greater values in that study (24). However, when applied to our data, the cut-off concentration of 2 $\mu\text{g/dL}$ or more is of little value in distinguishing CS from PCS. Although 99.6% of patients with CS were correctly identified, the specificity of this cut-off point was only 26.1%. This is not surprising, as patients with PCS have higher cortisol production than normal individuals.

The hypercortisolism of CS may be episodic (25–27). Hence, some patients with CS may have UFC levels within the normal range at the time of evaluation. This was noted in 22 of 221 (10%) of our CS patients, consistent with our earlier observations (28). Interestingly, a single midnight cortisol measurement correctly identified 20 of these patients, suggesting that it is a more sensitive test than UFC measurement for the diagnosis of early or episodic CS. Conversely, most patients with PCS have only mild elevation of UFC; at the time of the study only 2 of 23 had UFC greater than 200 $\mu\text{g/day}$. Thus, although consistent elevation of UFC greater than 200 $\mu\text{g/day}$ is suggestive of CS, additional test-

ing with serum cortisol values may be useful to further support the diagnosis.

Although a midnight serum cortisol value of 7.5 $\mu\text{g}/\text{dL}$ correctly diagnosed 225 of 234 patients with CS, it failed to detect 9 patients with CS. Six of these patients had consistently mild hypercortisolism (UFC, $<200 \mu\text{g}/\text{day}$), whereas 2 had episodic disease, with UFC below 90 $\mu\text{g}/\text{day}$ at the time of evaluation; 1 had UFC excretion between 330–523 $\mu\text{g}/\text{day}$. Therefore, as is the case with other diagnostic tests for CS, the midnight serum cortisol test may be falsely negative if the disease is mild or quiescent. As CS is a progressive disease, longer observation with repeated periodic testing is warranted in such cases, especially if clinical progression occurs. Unfortunately, we were forced to exclude 6 patients with failed TSS and negative pathology from this study because of inadequate follow-up. Thus, this study cannot assess the usefulness of midnight cortisol for the evaluation of such patients, who can pose a diagnostic dilemma (29).

What is the utility of midnight cortisol levels in the diagnosis of CS? We believe that this test is especially useful in patients who present with discordant clinical and biochemical results. Thus, a normal midnight cortisol value in a patient with few clinical features and elevated UFC would lend support to the diagnosis of PCS, whereas an elevated value might provoke continued evaluation of CS. The inconvenience associated with obtaining a midnight cortisol sample in a physician's office or other out-patient setting mitigates against any recommendation that midnight sampling be used as a routine screening test. However, it may be an extremely useful adjunct in patients who pose a diagnostic dilemma. The importance of prolonged clinical and laboratory evaluation of these difficult patients with multiple modalities, including midnight cortisol, cannot be overemphasized.

Acknowledgment

The authors acknowledge the contribution of Dr. Lynn D. Loriaux in the design of the study.

References

- Orth DN. 1995 Cushing's syndrome [published erratum appears in *N Engl J Med* 1995 Jun 1;332(22):1527]. *N Engl J Med*. 332:791–803.
- Gold PW, Loriaux DL, Roy A, et al. 1986 Responses to corticotropin-releasing hormone in the hypercortisolism of depression and Cushing's disease. Pathophysiologic and diagnostic implications. *N Engl J Med*. 314:1329–1335.
- Lamberts SW, Klijn JG, de Jong FH, Birkenhager JC. 1979 Hormone secretion in alcohol-induced pseudo-Cushing's syndrome. Differential diagnosis with Cushing disease. *JAMA*. 242:1640–1643.
- Wand GS, Dobs AS. 1991 Alterations in the hypothalamic-pituitary-adrenal axis in actively drinking alcoholics. *J Clin Endocrinol Metab*. 72:1290–1295.
- Smith MA, Kling MA, Whitfield HJ, et al. 1989 Corticotropin-releasing hormone: from endocrinology to psychobiology. *Horm Res*. 31:66–71.
- Tsigos C, Young RJ, White A. 1993 Diabetic neuropathy is associated with increased activity of the hypothalamic-pituitary-adrenal axis. *J Clin Endocrinol Metab*. 76:554–558.
- Roy MS, Roy A, Gallucci WT, et al. 1993 The ovine corticotropin-releasing hormone-stimulation test in type I diabetic patients and controls: suggestion of mild chronic hypercortisolism. *Metabolism*. 42:696–700.
- Yanovski JA, Cutler GBJ, Chrousos GP, Nieman LK. 1993 Corticotropin-releasing hormone stimulation following low-dose dexamethasone administration. A new test to distinguish Cushing's syndrome from pseudo-Cushing's states. *JAMA*. 269:2232–2238.
- Tsigos C, Papanicolaou DA, Chrousos GP. 1995 Advances in the diagnosis and treatment of Cushing's syndrome. *Bailliere Clin Endocrinol Metab*. 9:315–336.
- Pavlatos FC, Smilo RP, Forsham PH. 1965 A rapid screening test for Cushing's syndrome. *JAMA*. 193:720–723.
- Crapo L. 1979 Cushing's syndrome: a review of diagnostic tests. *Metabolism*. 28:955–977.
- Murphy BE. 1991 Steroids and depression. *J Steroid Biochem Mol Biol*. 38:537–559.
- Meikle AW, Lagerquist LG, Tyler FH. 1975 Apparently normal pituitary-adrenal suppressibility in Cushing's syndrome: dexamethasone metabolism and plasma levels. *J Lab Clin Med*. 86:472–478.
- Terzolo M, Borretta G, Ali A, et al. 1995 Misdiagnosis of Cushing's syndrome in a patient receiving rifampicin therapy for tuberculosis [published erratum appears in *Horm Metab Res* 1995 May;27(5):261]. *Horm Metab Res*. 27:148–150.
- Yanovski JA, Cutler Jr GB, Chrousos GP, Nieman LK. Prospective evaluation of the dexamethasone-suppressed corticotropin-releasing hormone test in the differential diagnosis of Cushing syndrome and pseudo-Cushing states. *Proc of the 77th Annual Meet of The Endocrine Soc*. 1995; 99.
- Krieger DT, Allen W, Rizzo F, Krieger HP. 1971 Characterization of the normal temporal pattern of plasma corticosteroid levels. *J Clin Endocrinol Metab*. 32:266–284.
- Pirich K, Vierhapper H. 1988 24-hour serum concentration profile of cortisol in patients with Cushing's disease. *Exp Clin Endocrinol*. 92:275–279.
- Follenius M, Brandenberger G, Hietter B. 1982 Diurnal cortisol peaks and their relationships to meals. *J Clin Endocrinol Metab*. 55:757–761.
- Swets JA. 1988 Measuring the accuracy of diagnostic systems. *Science*. 240:1285–1293.
- Friedman TC, Yanovski JA. 1995 Morning plasma free cortisol: inability to distinguish patients with mild Cushing syndrome from patients with pseudo-Cushing states. *J Endocrinol Invest*. 18:696–701.
- Glass AR, Zavadil III AP, Halberg F, Cornelissen G, Schaaf M. 1984 Circadian rhythm of serum cortisol in Cushing's disease. *J Clin Endocrinol Metab*. 59:161–165.
- Tourniaire J, Chalendar D, Rebattu B, et al. 1986 The 24-h cortisol secretory pattern in Cushing's syndrome. *Acta Endocrinol (Copenh)*. 112:230–237.
- Shimatsu A, Kato Y, Sakami H, Nakai Y, Imura H. 1988 Phase-shifted diurnal rhythm of cortisol secretion in a patient with Cushing's syndrome due to adrenocortical adenoma. *Endocrinol Jpn*. 35:261–266.
- Newell-Price J, Trainer P, Perry L, Wass J, Grossman A, Besser M. 1995 A single sleeping midnight cortisol has 100% sensitivity for the diagnosis of Cushing's syndrome. *Clin Endocrinol (Oxf)*. 43:545–550.
- Atkinson AB, Kennedy AL, Carson DJ, Hadden DR, Weaver JA, Sheridan B. 1985 Five cases of cyclical Cushing's syndrome. *Br Med J (Clin Res Ed)*. 291:1453–1457.
- Streeten DHP, Anderson Jr GH, Dalakos T, Joachimpillai AD. 1997 Intermittent hypercortisolism: a disorder strikingly prevalent after hypophysial surgical procedures. *Endocr Pract*. 3:123–129.
- Velkeniers B, Beckers A, Stevenaert A, Smits J, Finne E, Vanhaelst L. 1991 Cyclical Cushing's disease. A case report. *Pathol Res Pract*. 187:603–607.
- Nieman LK, Cutler Jr GB. The sensitivity of the urine free cortisol measurement as a screening test for Cushing's syndrome. *Proc of the 72nd Annual Meet of The Endocrine Soc*. 1990; 230.
- Atkinson AB, McCance DR, Kennedy L, Sheridan B. 1992 Cyclical Cushing's syndrome first diagnosed after pituitary surgery: a trap for the unwary. *Clin Endocrinol (Oxf)*. 36:297–299.