# The Desmopressin Test Predicts Better Than Basal Cortisol the Long-Term Surgical Outcome of Cushing's Disease

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**Context:** Cushing's disease (CD) has a significant relapse rate after successful transsphenoidal surgery (TSS). Many CD patients respond aberrantly to the desmopressin test (DT). Disappearance of this response after surgery may suggest complete removal of abnormal corticotrophs and a lower possibility of recurrence.

**Objective:** The utility of postoperative DT to predict long-term outcome compared to the widely used postoperative cortisol level.

**Design:** Retrospective analysis.

**Setting:** Tertiary hospital.

**Patients:** Seventy-three patients underwent TSS and postoperative DT; 51 had sustained remission, defined as normal dexamethasone suppression and urinary free cortisol at 6 months. After excluding 12 patients with short follow-up, negative or no preoperative DT, we analyzed 39 patients.

**Intervention(s):** Measurements of morning cortisol at 1–2 weeks and DT within 6 months after TSS.

**Main Outcome Measure(s):** Recurrence or remission at latest follow-up.

**Results:** Mean follow-up was 63 $\pm$ 50 months. Recurrence occurred in seven patients. In logistic regression analysis, postoperative cortisol levels were not associated with remission. Apart from the percentage increment of cortisol, all other DT criteria (peak cortisol, peak ACTH, absolute cortisol increment [$\Delta$Cort], absolute ACTH change, and percentage absolute ACTH change) were significant predictors of outcome. In receiver operating characteristic analysis, the $\Delta$Cort had the best diagnostic performance. $\Delta$Cort $<$7.4 $\mu$g/dL had a sensitivity of 97% to detect remission. Comparison of Kaplan-Meier curves showed that $\Delta$Cort $<$7.4 $\mu$g/dL was associated with remission, whereas $\Delta$Cort $\geq$7.4 $\mu$g/dL had a hazard ratio of recurrence of 24.7 (95% confidence interval, 10.6–448.5) at 60 months (median).

**Conclusion:** Loss of desmopressin response indicates favorable prognosis and, if used in addition to basal cortisol levels, improves the accuracy of the postoperative assessment of CD. **(*J Clin Endocrinol Metab* 101: 4878–4885, 2016)**

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Abbreviations: $ACTH_0$, ACTH at time 0; $ACTH_{15,30}$, ACTH at 15 or 30 minutes; $\Delta$ACTH, absolute ACTH change; AUC, area under the curve; CD, Cushing's disease; CI, confidence interval; $Cort_0$, cortisol at time 0; $Cort_{30,45}$, cortisol at 30 or 45 minutes; $\Delta$Cort, absolute cortisol change; DT, desmopressin test; GLM, general linear model; HPA, hypothalamic-pituitary-adrenal; NS, not significant; ROC, receiver operating characteristic; TSS, transsphenoidal surgery; UFC, urinary free cortisol.

Cushing's disease (CD) is a detrimental disease with high morbidity, decreased quality of life, and increased mortality (1, 2). Transsphenoidal surgery (TSS) remains the optimal therapeutic choice for patients with CD (3). The success rate of TSS, however, is far from ideal, with immediate remission rates ranging from 69 to 98% (3, 4). Even after initially successful TSS, the long-term outcome is further compromised by substantial late recurrence rates of 15–66% within 5 to 10 years (4, 5), mandating frequent lifelong postoperative follow-up and subsequent therapies.

Recognition of factors that can identify patients at high risk for recurrence or, conversely, those more likely to attain long-term remission will result in better consultation and planning of follow-up. So far, several factors have been proposed as predictors of long-term outcome (3, 4, 6). The most widely used criterion is a low early postoperative cortisol level. In a recent structured review and meta-analysis (7), many, but not all, studies validated it as a good predictor for long-term remission. However, a considerable number of patients with low early postoperative cortisol recur (8), and a small but significant number of patients achieve late remission (9). Other parameters studied as possible predictors include prolonged suppression of the hypothalamic-pituitary-adrenal (HPA) axis (6, 10), normal late night salivary cortisol (11), normal cortisol suppression by low-dose dexamethasone (12), and a postoperative CRH test (10, 13). None of these parameters, however, has been shown to be accurate enough in the prediction of long-term remission (3).

Desmopressin (1-deamino 8-D-arginine vasopressin) is a synthetic analog of vasopressin that is relatively specific for the renal V2 receptor. A significant number of corticotroph adenomas express this receptor aberrantly (14), and contrary to normal subjects, many patients with CD have paradoxical ACTH and cortisol responses to the desmopressin test (DT) (15–17). The rationale for using the DT postoperatively is based on the hypothesis that persistence of the desmopressin response indicates the presence of residual neoplastic corticotrophs, and hence an increased risk for relapse. In previous studies (18–24), the DT was performed at varying intervals after TSS and many different criteria were applied, resulting in a great variability in its diagnostic ability to predict long-term outcome.

In the present study, we aimed to evaluate whether the loss of a positive preoperative desmopressin responsiveness during the early postoperative period predicts long-term remission in patients with CD after successful TSS and to compare it with the most widely used and studied criterion of early postoperative basal morning cortisol measurement.

## Patients and Methods

At the Evangelismos Hospital, DT has been part of the standard preoperative, as well as postoperative, workup of CD since 1998. Informed consent was obtained from all patients. The institutional review board approved the retrieval of records. In this series, we analyzed the records of patients who underwent both preoperative DT and early (within 6 months after TSS) postoperative DT. Inclusion criteria were sustained postsurgical remission, positive preoperative desmopressin, and follow-up for more than 1 year. None of the subjects received medical treatment for CD preoperatively. Patients were investigated soon after surgery, at 3- to 6-month intervals initially, and then yearly in the long term.

The diagnosis of CD was based on the following biochemical criteria: nonsuppressed ACTH levels (>10 pg/mL), positive CRH test, more than a 50% decrease in cortisol after a high-dose dexamethasone suppression test, and positive bilateral inferior petrosal sinus sampling in cases with equivocal results. Confirmatory criteria were: positive histology for basophilic adenoma with positive immunostaining for ACTH; sustained clinical and laboratory remission, despite negative histology; or positive bilateral inferior petrosal sinus sampling.

We defined post-TSS remission as occurring when the following criteria were met: 1) patients with low serum cortisol levels <5 μg/dL early (1–2 weeks) after surgery were characterized as having early remission (3); 2) patients with higher cortisol levels (>5 μg/dL) early (1–2 weeks) after surgery but low or normal urinary free cortisol (UFC) levels and normal overnight dexamethasone suppression test (below 1.8 μg/dL) within 1 month from TSS were considered as having delayed remission (11); and 3) there were symptoms of adrenal insufficiency necessitating corticosteroid replacement or improvement in clinical features. All included patients had documented sustained remission at 6 months after surgery by demonstrating normal UFC and normal levels of cortisol suppression after dexamethasone suppression (<1.8 μg/dL).

Overall, 87 patients with CD were admitted to the Evangelismos Hospital during the study period. We identified 73 patients who underwent postoperative DT. Of those, 51 (70%) fulfilled sustained remission criteria, three had very short follow-up (<1 year), five had negative preoperative DT, and no preoperative DT was performed in four patients. The reason for not performing the preoperative DT was that a patient was referred to our center postoperatively. Overall, 39 patients were eligible for inclusion. All studied patients were Caucasian. Histology was positive in 32 of the 39.

Recurrence was defined by the reappearance of clinical and biochemical hypercortisolism based on at least two of the following criteria: unsuppressed cortisol after a 1-mg dexamethasone suppression test (>1.8 μg/dL), increased midnight serum cortisol, and increased UFC levels (mean of two samples).

We performed the DT (desmopressin, 10 μg iv bolus; Ferring Pharmaceuticals Ltd.) as follows. After an overnight fast, an indwelling catheter was inserted at 8 AM with the subject remaining supine during the whole study period. At 8:30 AM (0 minutes), 10 μg desmopressin was given as an iv bolus injection. Blood samples for ACTH and cortisol measurements were obtained at −15, 0, 15, 30, 45, 60, 90, and 120 minutes. Blood pressure and heart rate were recorded during the study period, and restriction of fluids was advised for the rest of the day. No side effects were observed. The following parameters were considered: ACTH at

time 0 ($ACTH_0$), cortisol at time 0 ($Cort_0$), peak ACTH at 15 or 30 minutes (peak $ACTH_{15,30}$), peak cortisol at 30 or 45 minutes ($peakCort_{30,45}$), absolute ACTH change (ΔACTH = peak $ACTH_{15,30} - ACTH_0$), absolute cortisol change (ΔCort = $peakCort_{30,45} - Cort_0$), percentage of ACTH change (%ΔACTH = [ΔACTH/$ACTH_0$] ×100), and percentage of cortisol change (%ΔCort = [ΔCort/$Cort_0$] × 100). For the preoperative DT, an increment of cortisol (%ΔCort) more than 20% and/or of ACTH (%ΔACTH) more than 50% over the baseline values was considered to represent a positive response (16).

### Measurements

Serum total cortisol was assayed using an automated immunochemiluminescence assay method with the ADVIA Centaur CP immunoassay system (ACS, 180 cortisol assay; Bayer) (lower detection limit of the assay, 0.19 μg/dL; intra-assay coefficient of variation, 8.0% for 5.43 μg/dL, 6.4% for 14.86 μg/dL, and 9.2% for 31.78 μg/dL). The same immunochemiluminescence assay was used to measure UFC after dichloromethane extraction. We measured ACTH by immunoradiometric assay (Cis Bio International) with a lower detection limit of 2 pg/mL.

### Statistics

We used the IBM SPSS statistical package, version 20 (IBM Software Group) and GraphPad Prism, version 5.0 (GraphPad Software). We compared receiver operating characteristic (ROC) curves with MedCalc version 16.4.3. Data are presented as the mean value ± SD of the mean. We checked normality with the Shapiro-Wilk test. Where necessary, logarithmic transformation was applied to normalize data before using parametric tests. For group comparisons we used the unpaired *t* test, Mann-Whitney *U* test, $\chi^2$ analysis, or Fisher's exact test where appropriate. We chose a general linear model (GLM) repeated-measures analysis to examine changes of cortisol and ACTH during the DT and identify differences between groups. We constructed ROC curves to examine the diagnostic test performance, which is the ability to discriminate between patients who will recur vs those who will not. Sensitivity against 100% specificity was plotted at each level, and the area under the curve (AUC) was computed by the nonparametric Wilcoxon statistic. The AUC represents the probability of correctly identifying patients who will recur. We compared ROC curves with MedCalc version 16.4.3. We calculated odds ratios for recurrence for each ROC-derived cutoff with logistic regression analysis. Kaplan-Meier recurrence-free curves were plotted for analysis with the log-rank and Gehan-Breslow-Wilcoxon tests. We considered differences significant at $P < .05$.

## Results

Of all patients in initial remission, six patients were considered as having delayed remission. Recurrence occurred in seven patients, five with early remission and two with delayed remission, at a median time of 56 (25–75% percentiles, 16–88; range, 12–109) months after TSS. The median follow-up of those who did not recur was 49 (25–75% percentiles, 29–88; range, 12–199) months. Patients with prolonged remission compared to the seven patients who showed recurrence did not differ in demographic and hormonal characteristics at diagnosis or in the time until recurrence or last visit (Table 1).

### Preoperative desmopressin

GLM repeated-measures analysis showed that ACTH and cortisol levels rose significantly ($P < .001$ for ACTH and $P < .001$ for cortisol) after the administration of desmopressin. There were no differences in the levels of cortisol and ACTH during the DT between patients who recurred and those who did not ($P = .51$ for cortisol and $P = .32$ for ACTH) (Figure 1, A and B); the interaction, however, was significant, indicating a different pattern of change over time, as demonstrated in Figure 1A.

**Table 1.** Preoperative Characteristics of the Patients and Comparison Between Patients With Recurrence and Those in Remission

| | All | Long-Term Remission | Recurrence | *P* Value |
|---|---|---|---|---|
| n | 39 | 32 | 7 | |
| Age at diagnosis, y | 43.4 ± 11.6 | 43.6 ± 11.3 | 42.7 ± 13.6 | NS |
| Gender | 32 F/7 M | 26 F/6 M | 6 F/1 M | NS |
| BMI, $kg/m^2$ | 31.8 ± 6.1 | 30.9 ± 5.5 | 35.5 ± 7.6 | NS |
| Hypertension, Y/N | 19/10 | 16/6 | 3/4 | NS |
| Diabetes, Y/N | 16/23 | 12/20 | 4/3 | NS |
| Osteoporosis, Y/N | 10/28 | 8/13 | 2/5 | NS |
| Dyslipidemia, Y/N | 22/17 | 21/11 | 1/6 | .03 |
| Morning cortisol, μg/dL | 26.9 ± 10.3 | 26.3 ± 10.9 | 29.7 ± 6.6 | NS |
| Morning ACTH, pg/mL | 60.5 ± 38.8 | 59.6 ± 37 | 64.7 ± 50.9 | NS |
| Midnight serum cortisol, μg/dL | 22.1 ± 9.7 | 22.4 ± 10.0 | 21.0 ± 8.7 | NS |
| Cortisol post-LDDST | 15.3 ± 8.7 | 14.5 ± 8.3 | 19.3 ± 11.2 | NS |
| 24-h UFC, μg/24 h | 349 ± 267 | 358.1 ± 282.9 | 309.3 ± 190.8 | NS |
| MRI | 8 M/24 μ/7 n | 5 M/20 μ/7 n | 3 M/4 μ | NS |
| Months until recurrence or last visit | 62.7 ± 49.7 | 65.1 ± 52.1 | 51.4 ± 37.9 | NS |

Abbreviations: F, females; M, males; Y, yes; N, no; LDDST, low-dose dexamethasone suppression test; MRI, magnetic resonance imaging; M, macroadenoma; μ, microadenoma; n, negative MRI. Values are expressed as mean ± SD.

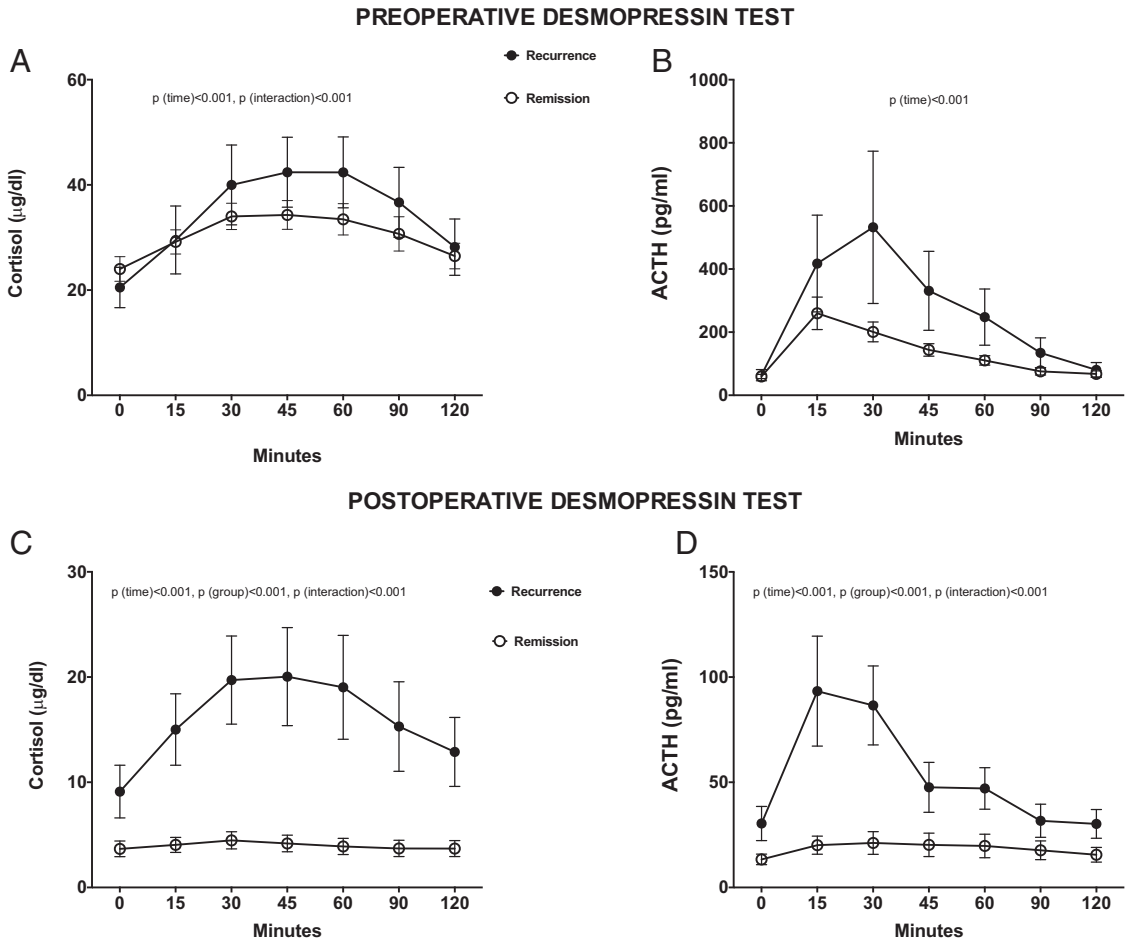


**Figure 1.** DT; cortisol (A) and ACTH (B) preoperatively, and cortisol (C) and ACTH (D) postoperatively in patients with and without recurrence.

## Postoperative desmopressin

The GLM repeated-measures analysis showed that both cortisol and ACTH increased significantly over time in the group of patients who recurred, but not in those who did not (both $P < .001$) (Figure 1, C and D). Cortisol levels increased significantly between 0 and 15 minutes and 15 and 30 minutes (time 0 vs 15, $P < .001$; and time 15 vs 30, $P < .001$), plateaued between times 30 and 45 ($P$ = not significant [NS]), and decreased significantly later on (time 60 vs 90, $P < .001$). ACTH levels rose significantly between times 0 and 15, plateaued between times 15 and 30, and then decreased (time 0 vs 15, $P < .001$; time 15 vs 30, $P$ = NS; and time 30 vs 45, $P < .001$). Table 2 shows the comparison between patients with recurrence and those in remission with regard to their early postoperative cortisol levels, as well as the biochemical parameters during DT.

## Comparison between early postoperative cortisol and DT

The difference in early postoperative cortisol levels between patients in long-term remission and patients who recurred was marginally insignificant ($P = .06$), and univariate logistic regression analysis showed that early postoperative cortisol levels could not predict the long-term outcome (Table 3). Recurrence occurred in 11.5% of patients with early postoperative cortisol levels <2 $\mu$g/dL and in 31% of those with higher cortisol levels.

With regard to DT, patients who recurred had higher peak $ACTH_{15,30}$, peak $Cort_{30,45}$, ΔACTH, ΔCort, %ΔACTH, and %ΔCort during the postoperative DT (Table 2). In univariate logistic regression analysis, all parameters except %ΔCort were significant predictors of the outcome (Table 3). $Cort_0$ and $ACTH_0$ were higher in patients who recurred (Table 2). In multivariate logistic analysis, ΔCort and ΔACTH remained significant after adjusting for $Cort_0$ and $ACTH_0$, respectively.

For the early postoperative cortisol level, the AUC of the ROC curve was not significant, in contrast to the parameters derived from the DT (Table 4). Pairwise comparisons of the ROC curves showed that ΔCort performed significantly better compared to both the early postoperative cortisol (difference between areas, 0.207 ± 0.10; 95% confidence interval [CI], 0.01–0.40; $P = .04$) and the %ΔCort ROC curves (difference between areas, 0.111 ± 0.05; 95% CI, 0.01–0.21; $P = .04$).

**Table 2.** Comparison Between Patients With Recurrence and Those in Remission With Regard to Their Early Postoperative Cortisol Levels, as Well as the Biochemical Parameters During Postoperative DT

| | Long-Term Remission | Recurrence | *P* Value |
|---|---|---|---|
| n | 32 | 7 | |
| Early postoperative cortisol | 2.4 ± 3.0 | 5.8 ± 7.8 | .06 |
| DT | | | |
| Timing of desmopressin after TSS (months) | 1.7 ± 2.3 | 2.7 ± 3.0 | NS |
| $Cort_0$ | 3.7 ± 4.2 | 9.1 ± 6.6 | .01 |
| Peak $Cort_{30,45}$ | 4.6 ± 4.7 | 20.7 ± 12.2 | .01 |
| ΔCort | 0.9 ± 3.0 | 11.6 ± 7.9 | .01 |
| %ΔCort | 54.0 ± 135.5 | 148.2 ± 107.6 | .09 |
| $ACTH_0$ | 13.4 ± 13.8 | 30.4 ± 21.4 | .01 |
| Peak $ACTH_{15,30}$ | 22.9 ± 29.8 | 101.8 ± 65.5 | .02 |
| ΔACTH | 9.8 ± 19.2 | 71.4 ± 45.9 | .01 |
| %ΔACTH | 56.7 ± 73.3 | 236.1 ± 125.1 | <.001 |

Abbreviations: $ACTH_0$, ACTH at time 0; $Cort_0$, cortisol at time 0; peak $ACTH_{15,30}$, peak ACTH at 15 or 30 min; peak $Cort_{30,45}$, peak cortisol at 30 or 45 min; ΔACTH = peak $ACTH_{15,30}$ − $ACTH_0$; ΔCort = peak $Cort_{30,45}$ − $Cort_0$; %ΔACTH = (ΔACTH/$ACTH_0$) × 100; %ΔCort = (ΔCort/$Cort_0$) ×100. Values are expressed as mean ± SD.

### Postoperative DT response criteria

The cutoffs that are associated with the best combination of sensitivity and specificity are shown in Table 4. Overall, ΔCort <7.4 μg/dL has comparable specificity (86%) to other criteria but better sensitivity (97%) and the highest odds ratio for remission of 186 (95% CI, 10–340.2; $P < .001$). Accordingly, Kaplan-Meier analysis showed that patients with ΔCort <7.4 μg/dL remained in prolonged remission, whereas those with ΔCort ≥7.4 μg/dL had a hazard ratio for recurrence of 24.7 (95% CI, 10.6–445.7) at a median recurrence-free period of 60 months. Figure 2, A and B, presents the curves for groups according to early postoperative cortisol levels (Figure 2A) ($P$ = NS) and ΔCort (Figure 2B) ($P < .001$). Only one patient with ΔCort <7.4 μg/dL recurred; repeated DTs were negative until 3 years postoperatively (ΔCort, 9 μg/dL), preceding documented recurrence by 21 months. Also, one patient with ΔCort >7.4 μg/dL is still in remission after 72 months of follow-up. Among the six patients with delayed remission, DT was performed at the same time with the early postoperative cortisol in four; it was negative in two who remained in prolonged remission and positive in two who recurred later on.

**Table 3.** Univariate Logistic Regressions for the Ability of Several Parameters to Predict Recurrence

| | Odds Ratio | 95% CI | *P* |
|---|---|---|---|
| Early postoperative cortisol | 1.15 | 0.97–1.38 | NS |
| $Cort_0$ | 1.21 | 1.02–1.43 | .03 |
| Peak $Cort_{30,45}$ | 1.31 | 1.08–1.58 | <.01 |
| ΔCort | 1.53 | 1.15–2.04 | <.01 |
| %ΔCort | 1.00 | 0.99–1.01 | NS |
| $ACTH_0$ | 1.06 | 1.00–1.11 | .03 |
| Peak $ACTH_{15,30}$ | 1.04 | 1.01–1.06 | <.01 |
| ΔACTH | 1.06 | 1.02–1.10 | <.01 |
| %ΔACTH | 1.02 | 1.01–1.03 | <.01 |

Abbreviations: $ACTH_0$, ACTH at time 0; $Cort_0$, cortisol at time 0; peak $ACTH_{15,30}$, peak ACTH at 15 or 30 min; peak $Cort_{30,45}$, peak cortisol at 30 or 45 min; ΔACTH = peak $ACTH_{15,30}$ − $ACTH_0$; ΔCort = peak $Cort_{30,45}$ − $Cort_0$; %ΔACTH = (ΔACTH/$ACTH_0$) × 100; %ΔCort = (ΔCort/$Cort_0$) × 100.

## Discussion

The findings from the present study suggest that a postoperative DT is a useful tool to stratify the risk of recurrence in successfully operated patients with CD. In fact, the postoperative DT performed better compared to the traditional measurement of early postoperative cortisol levels, which in this study was of questionable value in predicting the risk of recurrence. We also confirmed previous observations that a number of patients display late remission. Disappearance of a positive preoperative response to desmopressin may facilitate recognition of this subgroup and avoid unnecessary second surgery in these patients.

CD remains a difficult-to-treat condition. TSS is the best therapeutic option and the only one that can restore normal HPA axis function (25). However, even after successful surgery, cure rates are compromised by a high relapse rate. In the current study, 18% of patients finally presented recurrence after up to 109 months of follow-up (median, 56 months), in agreement with previous reports of up to 25% of recurrence at 10 years (4, 6). Discriminating between patients at high risk and those at low risk for recurrence is crucial for management decisions and, particularly, in low-risk patients where a vigorous follow-up may cause unnecessary inconvenience and anxiety. Hence, a number of studies attempted to identify predictors of the long-term outcome of patients operated on successfully for CD, but with limited success (5–7).

The DT has gained special interest because normal pituitary corticotrophs do not express V2 receptors, and thus most normal subjects do not respond to desmopressin challenge (16, 17). Responsiveness to desmopressin denotes the presence of tumorous corticotrophs expressing aberrantly V2 receptors, indicating the presence of CD. In this regard, varying according to arbitrary chosen criteria, responsiveness of established cases of CD to desmopressin at the time of diagnosis is observed in about 75–85% (16, 17, 21, 22). Persistence of such a "paradoxical" response

**Table 4.** ROC Analysis, Sensitivity, Specificity, and Odds Ratios of the Derived Thresholds for Predicting Remission

| Parameter | AUC ± SE | 95% CI | *P* | Cutoff | Specificity, % | Sensitivity, % | Odds Ratio (95% CI) | *P* |
|---|---|---|---|---|---|---|---|---|
| Early postoperative cortisol, μg/dl | 0.73 ± 0.1 | 0.55–0.91 | .06 | <1.3 | 86 | 50 | 6.0 (0.65–55.7) | NS |
| | | | | <2.0 | 57 | 72 | 3.4 (0.6–18.4) | NS |
| $Cort_0$, μg/dl | 0.80 ± 0.1 | 0.64–0.96 | .02 | <3.4 | 86 | 60 | 11.5 (1.2–107.5) | .03 |
| Peak $Cort_{30,45}$, μg/dl | 0.91 ± 0.1 | 0.78–1.00 | <.001 | <11.4 | 86 | 91 | 58.0 (5.1–657.4) | .001 |
| ΔCort, μg/dl | 0.94 ± 0.1 | 0.83–1.00 | <.001 | <7.4 | 86 | 97 | 186.0 (10.2–3402.1) | .001 |
| %ΔCort, % | 0.84 ± 0.1 | 0.71–0.96 | .01 | <62.5 | 86 | 81 | 10.8 (1.7–69.9) | .01 |
| $ACTH_0$, pg/ml | 0.80 ± 0.1 | 0.64–0.96 | .02 | <16.6 | 86 | 77 | 18.9 (1.9–184.6) | .01 |
| Peak $ACTH_{15,30}$, pg/ml | 0.88 ± 0.1 | 0.73–1.00 | <.01 | <54 | 86 | 90 | 56.0 (4.9–635.4) | .001 |
| ΔACTH, pg/ml | 0.92 ± 0.1 | 0.81–1.00 | .001 | <52 | 71 | 97 | 70.0 (5.3–925.8) | .001 |
| %ΔACTH, % | 0.91 ± 0.1 | 0.80–1.00 | .001 | <111 | 86 | 83 | 28.8 (2.8–294.8) | .01 |

Abbreviations: $ACTH_0$, ACTH at time 0; $Cort_0$, cortisol at time 0; peak $ACTH_{15,30}$, peak ACTH at 15 or 30 min; peak $Cort_{30,45}$, peak cortisol at 30 or 45 min; ΔACTH = peak $ACTH_{15,30}$ − $ACTH_0$; ΔCort = peak$Cort_{30,45}$ − $Cort_0$; %ΔACTH = (ΔACTH/$ACTH_0$) × 100; %ΔCort = (ΔCort/$Cort_0$) × 100.

postoperatively indicates the residual presence of abnormal corticotrophs, providing some ground for the use of DT to predict long-term outcome. To our knowledge, six independent studies (18–24) and another from one of the groups including a number of overlapping cases (21, 22) have assessed the usefulness of DT in this regard. In these studies the reported sensitivities and specificities to detect recurrence varied greatly from 45 to 100% and from 57 to 100%, respectively. The main reasons for these discrepancies are the arbitrary definition of response and the different applied criteria for remission and recurrence that may misclassify several patients. Importantly, there are no data on the preoperative response to desmopressin in three studies (18, 20, 24), and it is likely that nonresponders may have been included, thus compromising the ability of the test to predict recurrence. In our study, one patient recurred despite having a clearly negative postoperative DT. In this patient, repeated DTs are available; the test became positive at 3 years after surgery, preceding documented recurrence by 21 months. We believe that this patient was initially "cured" but experienced a true recurrence, whereas the other recurrent cases represent persistence of pathological corticotrophs that evolved over time and led to clinical reappearance of CD. In this context, as an early marker of recurrence during follow-up, the DT has been evaluated in only one study including a small number of patients. In our cohort, repeated DT was available in a subset of patients. Nevertheless, our study focused primarily on the accuracy of a single early postoperative DT, not as an early detector of recurrence but as a predictor of long-term outcome, allowing an initial stratification of patients according to their ultimate risk of recurrence.

Regarding the specificity of the test to predict recurrence, it has been argued that normal subjects may also respond to a desmopressin challenge. In fact, we have previously reported (16) that a small proportion, around 15%, of normal obese subjects may display percentage increases of their cortisol and ACTH that overlap with those observed in patients with CD. Some of these cases were documented to relate to stress induced from the pro-

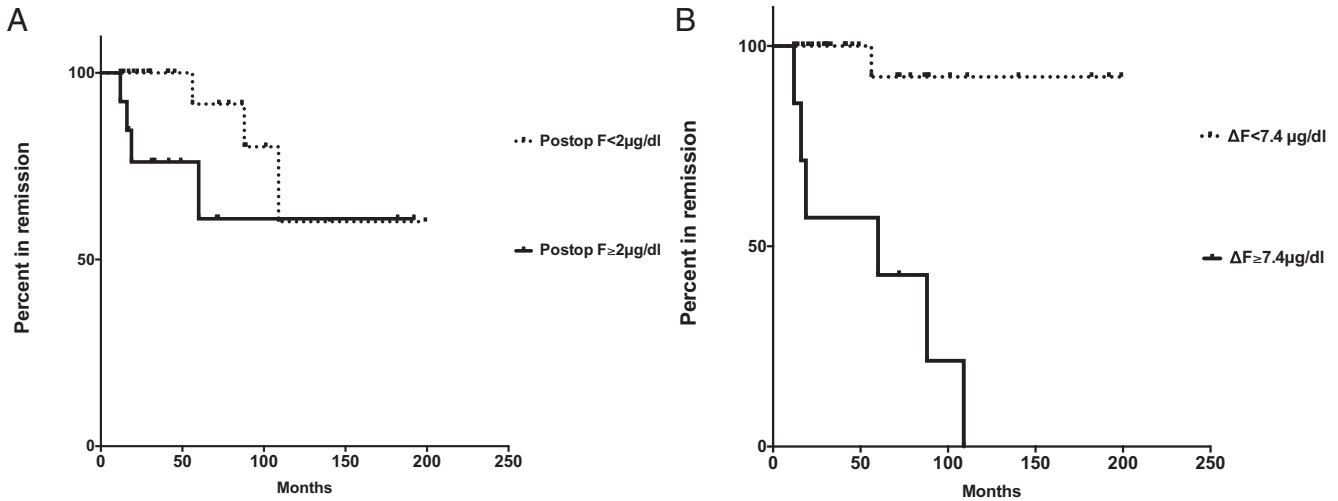


**Figure 2.** Kaplan-Meier curves according to their early postoperative cortisol (Postop F) levels (A) and the postoperative response to desmopressin (ΔF) (B).

cedure. Based on this, it has been suggested that concomitant dexamethasone suppression may increase the specificity of the DT. Thus, a modified protocol for the DT has been proposed that involves dexamethasone administration before performing the test (the so-called coupled dexamethasone-desmopressin test) (26). However, the case of patients with CD in the early postoperative period is somewhat different because in most of these patients, the HPA axis is already suppressed, and thus the need for further suppression by adding dexamethasone is not that crucial. In fact, one study reported similar performance of the two tests in the immediate postoperative period (20). The coupled dexamethasone-desmopressin test, however, may be more relevant as a tool to detect recurrence earlier than the conventional hormonal tests, as suggested by two studies (20, 26).

The diagnostic performance of the DT may also be compromised when response criteria based on percentage increments of cortisol are implemented. This is particularly relevant in the postoperative period, when cortisol levels are usually low and even small variations may result in percentage increases exceeding the widely used threshold of 20%, thus providing erroneously "positive" results. In fact, this is reflected in our results where the AUC of the ROC curve of the percentage increment of cortisol was significantly worse than that of the absolute increment of cortisol. In this line, previous studies that applied criteria based upon percentage increases (20–22) reported worse discriminatory ability of the postoperative DT, as opposed to studies that used absolute increments (19, 23). Of note, the proposed cutoff of ΔCort (7 and 7.3 μg/dL) in the latter studies is similar to that derived from our analysis, strengthening the validity of our results.

In previous studies, the measurement of early postoperative cortisol levels was the most frequently evaluated measure in predicting recurrence in CD patients, and it is widely incorporated into current clinical practice. In a recent meta-analysis (7), many but not all studies validated a low postoperative cortisol level as a favorable factor for long-term remission. In our cohort, however, 11.5% of patients with early postoperative cortisol levels $< 2$ μg/dL recurred, whereas 69% of patients with higher levels remained in prolonged remission, indicating a low predictive accuracy. These data are in good agreement with previous studies (8, 27) reporting that even with stringent criteria regarding postoperative cortisol levels (<1.8–2.0 μg/dL), 7–11.5% of the patients show a late relapse (8, 27). On the other hand, the presence of delayed remission in a number of patients with CD reduces the ability of the postoperative cortisol level to predict long-term remission (9). Identifying the patients that will display late remission is especially important for decisions regarding the need and timing of additional therapy. Although the number of cases is small, our analysis suggests that DT is more accurate in identifying remission in patients not achieving early hypocortisolism. Thus, patients who may especially benefit from the postoperative DT are those who do not have low cortisol levels within the first postoperative days but in whom the DT becomes negative. These patients may undergo late remission, and a watchful waiting approach may save them from unnecessary repeat surgery. However, persistence of the desmopressin response cannot exclude the possibility of late remission; herein it may indicate higher probability of late recurrence.

A potential limitation of our study is the relatively small, albeit comparable to other similar studies, number of CD patients and in particular the low number of patients with recurrence. This is because it is a single-center study including carefully selected patients, which at the same time constitutes the strength of this study. We chose only patients with a clearly positive preoperative DT, applied strict criteria for remission, sustained for at least 6 months postoperatively, and we also included only patients with a relatively prolonged follow-up to reduce the chance of missing late recurrences. We cannot exclude, however, the fact that an even longer follow-up might result in detecting more recurrences, which might occur even 20 years after surgery. Another significant advantage of this study is that we aimed to define the criteria for postoperative response based on the long-term outcome of patients and did not use previous arbitrarily defined criteria, as discussed above.

Our study has several clinical implications. We confirmed previous reports that a number of patients display late and sustained remission, despite not having low early postoperative cortisol levels, a finding that has implications on decisions regarding early re-treatment. In this context, the disappearance of a response to desmopressin appears to better recognize patients at postoperative remission. Furthermore, the postoperative DT, in contrast to early postoperative cortisol levels, can better stratify patients according to their risk of late relapse, and it is particularly useful in detecting patients at low risk for recurrence. In this regard, it should be noted that patients with values not exceeding but close to the proposed cutoff should be regarded as an intermediate group and should be treated with caution.

In conclusion, our data support a complementary role of the DT, in the postoperative assessment, as well as the planning of a follow-up of CD with a better performance compared to early cortisol measurements. The maintenance or disappearance of the response to desmopressin postoperatively may be related to the persistence or complete removal of adenomatous corticotrophs, mandating

close surveillance or indicating a good long-term outcome, respectively.

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