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DEXAMETHASONE SUPPRESSION TEST IN DIAGNOSIS OF CUSHING'S SYNDROME

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In the diagnosis of endocrine disease suppression tests have proved useful for revealing minor and clarifying doubtful instances of glandular overactivity. The value of the thyroid suppression test as an aid to the diagnosis of thyrotoxicosis is already well established. In normal subjects the secretion of thyroid-stimulating hormone by the pituitary can be suppressed by the administration of thyroid hormone; while in thyrotoxicosis such suppression either fails to occur or is incomplete (Werner and Spooner, 1955). Suppression tests of adrenocortical function are less well established in clinical medicine, partly because of the lower incidence of adrenal disease, and partly because much of the early work was concerned with using such tests to try to distinguish between Cushing's syndrome due to adrenal tumours and that due to bilateral adrenal hyperplasia—for example, Jailer *et al.* (1954)—rather than to distinguish between patients with either type of Cushing's syndrome and those with normal adrenal function. In the same way as in the thyroid suppression test, suppression of normal adrenocortical function following the administration of exogenous corticosteroids occurs as a sequel to the suppression of adrenocorticotrophic hormone production by the pituitary.

In Cushing's syndrome the basic abnormality is an excessive secretion of cortisol, the secretion rate of which can be measured by isotopic means (Cope and Black, 1958). Among the urinary steroid measurements probably the next best method for revealing oversecretion is the measurement of free cortisol (Cope and Black, 1959), which is estimated after chromatography. The most widely used urinary steroid estimation is that of the basal excretion of 17-ketogenic steroids (17-KGS) or of 17-hydroxycorticosteroids (17-OHCS). Others have used the levels of plasma steroids for the diagnosis of Cushing's syndrome (Grumbach *et al.*, 1955). The measurements of the cortisol-secretion rate, urinary free

cortisol, or plasma steroids all require special techniques which are not within the scope of many hospital laboratories. The disadvantage of the measurement of the basal values of urinary 17-KGS is that there is a considerable overlap between values found in normals and those found in cases of Cushing's syndrome (Cope and Black, 1959). Liddle (1960) has described the use of an adrenal suppression test in the diagnosis of Cushing's syndrome in which he estimated the urinary 17-OHCS after the administration of corticosteroid and found that the degree of suppression of the urinary 17-OHCS by the second day of steroid administration provided a good distinction between normal subjects and cases of Cushing's syndrome. We have also assessed the adrenal suppression test in the diagnosis of Cushing's syndrome, and our results confirm his in finding it valuable for this purpose. Like Liddle, we have used as suppressing agent 16 α -methyl-9 α -fluoro-prednisolone (dexamethasone), and we have determined its effect on the urinary excretion of 17-ketogenic steroids (17-KGS) as well as 17-ketosteroids (17-KS), these estimations being available routine procedures in most hospital laboratories.

Method

The *standard test* consisted of the collection of two 24-hour urine specimens, after which dexamethasone was given for a week and two more 24-hour urines were collected on the sixth and seventh days. Toluene was used in the bottles to prevent bacterial overgrowth. The standard dose of dexamethasone used was 0.5 mg. six-hourly, but in some a higher dose of 1 mg. six-hourly was also used.

Subjects Studied.—(1) Nine normal subjects, who consisted of volunteer doctors and nurses and one patient convalescent from a non-endocrine condition; (2) seven patients suffering from idiopathic hirsutism and four with menstrual disorders; (3) twenty-four subjects finally diagnosed as cases of "simple" obesity; and (4) six patients with Cushing's syndrome. Three of the patients with menstrual disorders had irregular periods and infertility, for which no adrenal or ovarian cause could be found, and the fourth had a congenital absence of the vagina. Assessment of the degree of obesity of the obese patients by the Kemsley (1952) standards indicated a mean excess weight of 73% above the normal for subjects of the same age and sex. The six patients with Cushing's syndrome all had the typical clinical picture, and the diagnosis in four of them was confirmed by the estimation of the cortisol secretion rate (by Dr. C. L. Cope); in one of the other two patients the 24-hour urine cortisol level was measured and was raised (170 μ g./24 hours). Radiography in all these patients after perirenal oxygen insufflation did not reveal evidence of an adrenal tumour, and they were all thought to be suffering from bilateral adrenal hyperplasia. One patient (D. J.), whose clinical details have already been published (Joplin and Fraser, 1961), had ocular signs suggesting an invasive tumour of the pituitary. All the patients were treated by pituitary implantation of ¹⁹⁸Au (Joplin *et al.*, 1961).

Procedures.—The 17-KS and the 17-KGS were estimated by the methods of Norymberski (Norymberski *et al.*, 1953). Basal collections on two separate days were obtained from 21 of the subjects with hirsutism or menstrual irregularities or obesity. The mean day-to-day variation in 17-KGS excretion was 2.3 mg., with a range of 0–7 mg. The precision of the

measurements was calculated from determinations done in duplicate. The estimate of the S.D. (Snedecor, 1952) for the values of the basal 17-KS was 0.39 mg.; the estimate of the S.D. for the values of the total 17-KS obtained after oxidation was 1.3 mg. for the basal values and 0.54 mg. for the values obtained after suppression.

Results

1. In the normal group, on the sixth and seventh days of dexamethasone administration, the urinary 17-KGS excretion fell to a mean of less than 1 mg./24 hours. All the individual values were under 2 mg./24 hours on these days, with the single exception of a subject whose 17-KGS excretion on the sixth day of dexamethasone was less than 1 mg., whereas the excretion on the seventh day was 4 mg.; this result being possibly due to strenuous physical exertion on that day.

2. In the group of patients with idiopathic hirsutism or menstrual disorders all the values of urinary 17-KGS on the sixth and seventh days of dexamethasone were also under 2 mg./24 hours (mean less than 1 mg./24 hours).

3. Nineteen obese subjects were tested on the standard dose of 0.5 mg. dexamethasone six-hourly, and their mean urinary 17-KGS on the sixth and seventh days of steroid administration were 1.1 and 1.2 mg./24 hours respectively. In 13 of these subjects the values were 2 mg. or less, but there were six who excreted between 2 and 4 mg. of 17-KGS on the sixth and seventh days of dexamethasone. Because of these less complete suppressions eight obese subjects were tested on the higher dose of dexamethasone of 1 mg. six-hourly; after this dosage all of them had values of urinary 17-KGS of 2 mg. or less per 24 hours on the sixth and seventh days, with the exception of one subject who excreted 4 mg. of 17-KGS on the sixth day of dexamethasone, though the value for the seventh day was less than 1 mg.

4. The individual results of the dexamethasone suppression test in the patients with Cushing's syndrome are shown in the Table and the Chart. It will be seen that a much smaller degree of suppression was found in these patients than was seen in any of the other groups; the values for the 17-KGS on the fifth, sixth, and

Results of Dexamethasone Suppression Test

Subjects	No.	Fig. Ref.	Dose of Dexamethasone	17-KS (Means and Ranges)			17-KGS (Means and Ranges)				
				Basal	Day of Dexamethasone			Basal	Day of Dexamethasone		
					5	6	7		5	6	7
Normal	9		2	—	4.4 (0.7)	3.8 (0.8)	—	0.3 (0.1)	0.7 (0.4)		
Hirsutism or menstrual disorder	8		2	9.75 (5-17)	3.3 (1.6)	2.7 (0.5)	5.4 (2-12)	0.2 (0.1)	0.4 (0.1)		
	3		4	13.3 (8-20)	3.7 (1.5)	2.5 (2.3)	7.3 (4-12)	0.5 (0.1)	0.5 (0.1)		
Obese	19*		2	9.4 (3-20)	3.5 (0.7)	3.2 (0.10)	7.6 (3-17)	1.1 (0.4)	1.2 (0.4)		
	8†		4	9.7 (3-18)	4.2 (1-8)	4.4 (0-8)	8.2 (6-15)	1.4 (0.4)	0.3 (0.1)		
Cushing's syndrome (untreated)	M.C. 25 F (a)		1.5	25, 27	14	13	45, 44	19	21		
	M.H. 60 F (b)		4	21		14	36		22		
	S.A. 11 F (c)		4	10, 8		7	31, 19		11		
	L.S. 47 F (d)		4	34, 37		21	14		19		
	E.C. 49 F (e)		4	8, 13		6	6		12		
	D.J. 27‡ M (f)		2	26		12	5		11		

* Includes one patient tested twice. † Includes two patients previously tested on 2-mg. dose. ‡ Patient with pituitary tumour.

Results of the dexamethasone suppression test.

seventh days of dexamethasone being above 10 mg./day in all the patients except one who excreted 5 mg. on one day but 11 mg. on the other. These values were found despite the fact that in four of them the higher corticosteroid dose level of 1 mg. six-hourly had been used. Four of the patients have been reassessed after pituitary implantation. Two of them (S. A. and M. C.) have shown a complete clinical remission. They have both had dexamethasone suppression tests, using a dose of 1 mg. six-hourly, during which the urinary 17-KGS fell to 1 mg. or less on the sixth and seventh days of dexamethasone. The patient with the pituitary tumour (D. J.) showed a partial remission following pituitary implantation; on testing him 40 weeks after the implant, the values of the urinary 17-KGS on the sixth and seventh days of dexamethasone in a dose of 1 mg. six-hourly were 7 and 6 mg. The sixth patient (M. H.) sustained no improvement following her implant; a dexamethasone suppression test performed 30 weeks after it showed 17-KGS values of 16 and 17 mg. on the fifth and sixth days of dexamethasone in a dose of 0.5 mg. six-hourly.

It will thus be seen that dexamethasone in a dose of 0.5 mg. six-hourly has produced suppression of the urinary 17-KGS to under 2 mg. a day in the normal subjects, and in those with idiopathic hirsutism or menstrual disorders, and also in most of the subjects with simple obesity. The patients with Cushing's syndrome showed much higher levels of 17-KGS after dexamethasone, while in two of those who had remitted clinically after implantation of the pituitary the dexamethasone suppression test was then also normal.

Discussion

The design of the dexamethasone suppression test we have used is similar to that used by Liddle (1960). He used 0.5 mg. six-hourly as the standard dose, and estimated the urinary 17-OHCS after two days of dexamethasone. He found that the excretion of 17-OHCS had fallen to less than 2.5 mg. on the second day of dexamethasone in normal subjects. We have used the same standard dose of dexamethasone, but have prolonged the period of suppression to seven days. This was done arbitrarily, and Liddle's data indicate that shorter periods of suppression may be adequate. Jayle *et al.* (1960), however, state that a dose of 2 mg. of dexamethasone a day does not produce complete inhibition until 8 to 10 days, and prefer to use a dose of 3 mg. a day for a period of five days. Dexamethasone has been used as a suppressing agent because it is a powerful glucocorticoid which can be used in doses so small that its derivatives in the urine will not interfere with the estimation of urinary steroids.

In the form of suppression test we have used, normal subjects suppressed to under 2 mg. of 17-KGS a day on a dose of 0.5 mg. of dexamethasone six-hourly. The majority of obese subjects showed similar degrees of suppression after dexamethasone; among them, however, were a few who did not show such complete degrees of suppression, though this was never sufficient to cause confusion with the levels seen in the patients with Cushing's syndrome. Significantly increased basal corticosteroid excretions in obese subjects have been reported (Poisnick and DiRaimondo, 1956), and their adrenals may be slightly less easily suppressed than those of normals.

It has already been mentioned that Cope and Black (1959) have emphasized the considerable overlap into

the normal range of values obtained for the basal urinary 17-KGS in patients with Cushing's syndrome. Using this urinary steroid estimation, we have been concerned especially to assess the value of the dexamethasone suppression test as an aid in the diagnosis of the presence or absence of Cushing's syndrome, rather than in its use in the differential diagnosis of the lesion causing Cushing's syndrome. After dexamethasone the levels of 17-KGS were much higher in the patients with Cushing's syndrome than in the other groups of subjects. The patients all had florid Cushing's syndrome, and in four of them considerably raised basal levels of urinary 17-KGS were found. In one of them (D. J.), however, the basal levels were only slightly raised, and in another (E. C.) they were within the normal range. After dexamethasone they both had values of 17-KGS excretion that were obviously above the values found in normal subjects after corticosteroid administration, indicating that the suppression test provides a more clear-cut index of adrenocortical overactivity than do the basal values of 17-KGS.

Liddle states that the urinary 17-KS excretion does not fall as readily during the suppression test as does the excretion of 17-OHCS. Our results agree with him in showing this smaller suppression of the 17-KS. This applies to both males and females, whose results have been grouped together in the Table for the sake of clarity. Jayle *et al.* (1961), however, found lower values of 17-KS after dexamethasone, with a mean of 1.4 mg. of urinary 17-KS in normal females on the fifth day of dexamethasone administration in a dose of 3 mg./day.

Summary

Results of a dexamethasone suppression test for assessing adrenocortical function are described. Dexamethasone in a dose of 0.5 mg. six-hourly will produce almost complete suppression of 17-KGS excretion (to under 2 mg./day) by the sixth and seventh days of administration in normal subjects and patients with idiopathic hirsutism or menstrual disorders. Slightly less complete suppression is seen in patients with simple obesity, sometimes down to only 4 mg./day. Much less complete suppression is seen in Cushing's syndrome, usually to values of over 10 mg./day. Thus this simple procedure is useful for the diagnosis of Cushing's syndrome.

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