RESEARCH

A study of thyroid functions in patients with Cushing's syndrome: a single-center experience

Boni Xiang[1,*], Ran Tao[1,*], Xinhua Liu[1], Xiaoming Zhu[1], Min He[1], Zengyi Ma[2], Yehong Yang[1], Zhaoyun Zhang[1], Yiming Li[1], Zhenwei Yao[3], Yongfei Wang[2] and **Hongying Ye[1]**

[1]Department of Endocrinology and Metabolism, Huashan Hospital, Fudan University, Shanghai, China
[2]Department of Neurosurgery, Huashan Hospital, Fudan University, Shanghai, China
[3]Department of Radiology, Huashan Hospital, Fudan University, Shanghai, China

Correspondence should be addressed to H Ye: yehongying@huashan.org.cn

*(B Xiang and R Tao contributed equally to this work)

Abstract

Objective: The aim of this study was to evaluate thyroid functions in Cushing's syndrome (CS), the dynamic changes of thyroid hormones and antithyroid antibodies in Cushing's disease (CD) pre- and postoperatively.
Design and methods: This is a retrospective study enrolling 118 patients with CS (102 CD, 10 adrenal CS and 6 ectopic adrenocorticotropic syndrome (EAS)). Thyroid functions (thyroid-stimulation hormone (TSH), T3, free T3 (FT3), T4 and free T4 (FT4)) were measured in all CS at the time of diagnosis and in all CD 3 months after transsphenoidal pituitary tumor resection. Postoperative hormone monitoring within 3 months was conducted in 9 CD patients completing remission. Twenty-eight remitted CD patients experienced hormone and antithyroid antibody evaluation preoperatively and on the 3rd, 6th and 12th month after surgery.
Results: TSH, T3 and FT3 were below the reference range in 31%, 69% and 44% of the 118 CS patients. Remitted CD patients (81/102) had significantly higher TSH ($P = 0.000$), T3 ($P = 0.000$) and FT3 ($P = 0.000$) than those in the non-remission group (21/102). After remission of CD, TSH, T3 and FT3 showed a significant increase, with a few cases above the reference range. By 12 months, most CD patients' thyroid functions returned to normal. Thyroid hormones (including TSH, T3 and FT3) were negatively associated with serum cortisol levels both before and after surgery. No significant changes of antithyroid autoantibodies were observed.
Conclusions: TSH, T3 and FT3 are suppressed in endogenous hypercortisolemia. After remission of CD, TSH, T3 and FT3 increased significantly, even above the reference range, but returned to normal 1 year after surgery in most cases. Antithyroid antibodies did not change significantly after remission of CD.

Key Words
- Cushing's syndrome
- Cushing's disease
- cortisol
- thyroid hormones
- antithyroid antibodies

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Introduction

Cushing's syndrome (CS) comprises diverse manifestations resulting from chronic exposure to excess glucocorticoids. The incidence is 0.2–5.0 per million people per year. Approximately 80% of endogenous CS is adrenocorticotrophin (ACTH)-dependent (1), and 20% is ACTH-independent. Pituitary corticotroph adenoma (Cushing's disease (CD)) is the most common cause (2), followed by primary unilateral adrenal adenomas and ectopic adrenocorticotropic syndrome (EAS). The clinical presentation includes reddish purple striae,

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plethora, proximal muscle weakness and metabolic disorders (central obesity, hypertension, diabetes mellitus and dyslipidemia).

The inhibitory effect of hypercortisolemia on the hypothalamic–pituitary axis is most commonly characterized by menstrual abnormalities in adults and growth retardation in children. Besides, the thyroid hormone changes in hypercortisolemia have been reported since 1952. Fredrickson *et al.* described that massive doses of cortisone acetate decreased iodine-131 (I^{131}) accumulation in the thyroid of euthyroid patients (3). Dexamethasone administration (16 mg daily for 2.5 days) was reported to reduce the thyroid-stimulating hormone (TSH) and free T3 (FT3) secretion and blunted the TSH response to thyrotropin-releasing hormone (TRH) (4). Regarding endogenous hypercortisolemia, a study of three patients with adrenal CS showed similar results (5). Kuku reported that serum TSH response to TRH was impaired in eight patients with CD. The impairment was relieved after a pituitary implant of ^{198}Au (6). Primary cortisol deficiency was reported concomitant with high TSH and low FT3. After cortisone administration, TSH returned to normal. However, serum cortisol and TSH showed no significant correlation (7).

In 2013, Tamada *et al.* first reported 'hyperthyroidism' in two CS patients after surgery due to 'syndrome of inappropriate secretion of TSH' (SITSH) associated with the insufficient replacement of hydrocortisone (HC) (8). Free T3 and TSH were normalized after a HC dose increase (8). Then, the same group investigated the clinical course in eight CS patients and found that free T3 levels were above the reference range in 75% of patients up to 6 months after surgery, and all returned to normal within 1 year (9). They suggested that cured CS patients might develop high T3 or T4 with normal or elevated TSH (8).

Endogenous CS is an uncommon disease. There has not yet been a large sample in which the clinical course of thyroid hormone changes was studied before and after remission of endogenous CS. In clinical practice, because of the lack of knowledge about this condition, some CS patients' thyroid functions may be mistaken as evidence of hypothyroidism or hyperthyroidism after surgery and even led to the medication of antithyroid drugs.

The aim of our study was to evaluate the thyroid functions of patients with CS, compare the hormone levels in patients with different etiologies, and investigate the clinical course of fluctuations of thyroid hormones in patients with CD pre- and postoperatively.

Patients and methods

Patients

This study included 118 CS patients who were hospitalized in the Department of Endocrinology and Metabolism of Huashan Hospital between January 2013 and April 2016. They were diagnosed with either CD ($n=102$, median age: 36.0 years, range 13–63 years, female/male: 90/12), adrenal CS ($n=10$, median age: 36.5 years, range 21–57 years, female/male: 8/2) or EAS ($n=6$, median age: 49.0 years, range 22–72 years, female/male: 5/1). All subjects had a comprehensive clinical evaluation by the same group of endocrinology specialists and all patients with CD underwent primary resection at our institution by the same surgeon. None of the patients had a history of thyroid diseases.

Methods

Data on clinical findings, laboratory findings, imaging findings, treatment and outcome were obtained. All CD patients (microadenomas: 88/102, macroadenomas: 14/102) underwent transsphenoidal pituitary tumor resection (TSS) through a microscope or endoscope. Remission was defined as morning serum cortisol levels lower than 5.0 μg/dL within 7 days of selective tumor resection (10). The cortisone replacement began after remission, and cortisone dosage prescribed was based on the serum cortisol levels and the symptoms. Thyroid hormones (TSH, T3, FT3, T4 and free T4 (FT4)) and serum cortisol were measured in all 118 patients before treatment. In all 102 CD patients, thyroid hormones and serum cortisol were measured 3 months after surgery. The hormones were measured before surgery and 1 day, 2 weeks, 1 month, 2 months and 3 months after surgery in nine CD patients completing remission. In 28 remitted CD patients, the hormones and antithyroid autoantibodies were measured 3, 6 and 12 months after surgery.

Hormonal assays

Samples were obtained at 08:00 h before any medication. Serum thyroid hormones (TSH, T3, FT3, T4 and FT4) and antithyroid autoantibodies (TPO-Ab and ATG) were assessed by immunoenzymometric assay method (ADVIA Centaur®; Siemens Healthcare Diagnostics Inc.). Serum cortisol was measured by chemiluminescent enzyme immunoassay (cobas® e 602, Roche Diagnostics).

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Statistical analyses

Normal distributed continuous variables were expressed as mean values ± standard deviation (s.d.). *T*-test was used to compare means between groups. Simple linear regression coefficients and multiple regression analysis were used to examine the correlation among parameters. SPSS 20.0 (SPSS) was used. A two-tailed *P* value <0.05 was considered statistically significant.

Ethical approval

The study was approved by the ethics committee of Huashan Hospital attached to Fudan University (#KY2017-422). Consent has been obtained from each patient or subject after full explanation of the purpose and nature of all procedures used.

Results

At the time of diagnosis, TSH, T3, FT3, T4 and FT4 levels were below the reference range in 31%, 69%, 44%, 11% and 19% of 118 CS patients, respectively (Table 1). None of the patients had elevated thyroid hormones. In 33 (28%) patients, low TSH with low T3 or T4 was found. Regarding different etiologies, serum cortisol in patients with EAS was higher than that in adrenal CS (44.1 ± 17.1 μg/dL vs 30.1 ± 7.8 μg/dL, *P*=0.04) and CD (44.1 ± 17.1 μg/dL vs 29.4 ± 10.1 μg/dL, *P*=0.001). FT3 and FT4 in EAS were also the lowest, with significance when comparing with those in CD (FT3: 2.93 ± 0.76 nmol/L vs 3.68 ± 0.68 pmol/L, *P*=0.009; FT4: 11.4 ± 1.6 nmol/L vs 13.9 ± 2.8 pmol/L, *P*=0.035) (Table 2).

Three months after surgery, 81/102 CD patients developed remission. The median dose of cortisone acetate for post-surgery and 3rd-month replacement treatment were separately 50.0 mg/day (0~100 mg/day) and 25.0 mg/day (0~75.0 mg/day) in remission group. Twenty of the 81 remitted patients needed no more cortisone replacement on the third month. Postoperative TSH (2.42 ± 1.73 mIU/L vs 1.07 ± 0.82 mIU/L, *P*=0.000), T3 (2.09 ± 0.77 nmol/L vs 1.08 ± 0.31 nmol/L, *P*=0.000), FT3 (5.37 ± 1.37 pmol/L vs 3.70 ± 0.66 pmol/L, *P*=0.000), T4 (93.1 ± 23.3 nmol/L vs 80.2 ± 18.4 nmol/L, *P*=0.000) were higher than those before surgery (Fig. 1 and Table 3). TSH in 11.1% (9/81), T3 in 4.9% (4/81) and FT3 in 17.3% (14/81) were above the reference range 3 months after surgery in the remission group. However, FT4 slightly decreased after surgery (before surgery: 13.8 ± 2.8 pmol/L, 3 months after surgery: 12.9 ± 2.7 pmol/L, *P*=0.012). Moreover, elevated T3, FT3 and FT4 with nonsuppressed TSH were seen in 12/81 (14.8%) cases.

We investigated the clinical parameters between the remission and non-remission groups (Fig. 1 and Table 3). Before surgery, serum cortisol and thyroid hormones showed no difference between the two groups. However, 3 months after surgery, TSH (*P*=0.000), T3 (*P*=0.000) and FT3 (*P*=0.000) became significantly higher in the remission group. T4 of the two groups showed no difference (*P*=0.668). FT4 levels in the remission group were lower than those in the non-remission group (12.9 ± 2.7 pmol/L vs 14.8 ± 2.7 pmol/L, *P*=0.004).

Correlation analyses between levels of serum cortisol and thyroid hormones were conducted in the 112 CS patients before surgery and the 102 CD patients 3 months after surgery. Significant negative correlations between thyroid hormones and serum cortisol both before and after surgery were found (Table 4). We also analyzed the correlations between tumor size and preoperative serum cortisol/thyroid functions in all the CD patients (*n*=102), but the association turned out insignificant. Besides, the correlation among several parameters, including thyroid hormones, serum cortisol levels, age, gender, BMI and tumor size were analyzed using simple linear regression analysis (Table 5). It revealed marked negative correlations between serum cortisol levels and thyroid function parameters including TSH (*P*=0.001), T3 (*P*=0.000) and FT3 (*P*=0.000) before surgery. In addition, BMI was positively associated with T3 (*P*=0.005) and FT3 (*P*=0.047) and age were inversely associated with FT3 (*P*=0.015) and FT4 (*P*=0.025). However, after surgery, the only variable significantly affecting thyroid hormones was serum cortisol (in postoperative analyses, the variable 'tumor size' was excluded).

For the nine remitted patients whose thyroid functions were closely monitored within 3 months, the

Table 1 Characteristics of patients with Cushing's syndrome before treatment.

	Cushing's syndrome	Reference range
Number	118	/
Age, years	36.7 (13.0–72.0)	/
Female/male	103/15	/
Serum cortisol (μg/dL)	27.9 (11.7–63.4)	/
TSH (mIU/L)	0.75 (0.03–4.55)	0.550~4.780
T3 (nmol/L)	1.02 (0.43–3.26)	1.23~3.39
T4 (nmol/L)	75.9 (25.8–131.2)	54.0~174.0
FT3 (pmol/L)	3.58 (1.27–5.33)	3.50~6.50
FT4 (pmol/L)	13.21 (6.36–21.82)	11.5~22.7

Data are median (minimum–maximum).

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Table 2 Comparative study between CD, adrenal adenoma and EAS.

	CD	Adrenal adenoma	EAS	Reference range
n	102	10	6	/
Age (years)	36.0 (13.0, 63.0)	36.5 (21.0, 57.0)	49.0 (22.0, 72.0)	/
Male/female	12/90	2/8	1/5	/
Serum cortisol (μg/dL)	29.4 ± 10.1	30.1 ± 7.8	44.1 ± 17.1[a]	/
TSH (mIU/L)	1.07 ± 0.82	0.65 ± 0.51	0.70 ± 0.79	0.55~4.78
T3 (nmol/L)	1.10 ± 0.38	1.12 ± 0.69	0.85 ± 0.31	1.23~3.39
T4 (nmol/L)	79.0 ± 19.0	72.6 ± 15.4	68.4 ± 35.1	54.0~174.0
FT3 (pmol/L)	3.68 ± 0.68[b]	3.32 ± 0.53	2.93 ± 0.76[b]	3.50~6.50
FT4 (pmol/L)	13.9 ± 2.8[b]	12.6 ± 2.6	11.4 ± 1.6[b]	11.5~22.7

[a]Serum cortisol of EAS was significantly higher than that of adrenocortical adenoma ($P = 0.04$) and CD ($P = 0.001$). [b]FT3 ($P = 0.009$) and FT4 ($P = 0.035$) in EAS were significantly lower than those in CD.

median dose of cortisone acetate for post-surgery, second week, first month, second month and third-month replacement treatment were 50.0 mg/day (0~75.0 mg/day), 50.0 mg/day (0~50.0 mg/day), 25.0 mg/day (0~50.0 mg/day), 25.0 mg/day (0~50.0 mg/day) and 25.0 mg/day (0~50.0 mg/day). Two of the nine patients received no cortisone replacement after surgery (Fig. 2). Among them, we found that TSH started to rise since the first day after surgery (the increase became statistically remarkable since the second week and the significance lasted till the third month). FT3 and T3 became lower on the first day and then rose up markedly. The changes of T4 and FT4

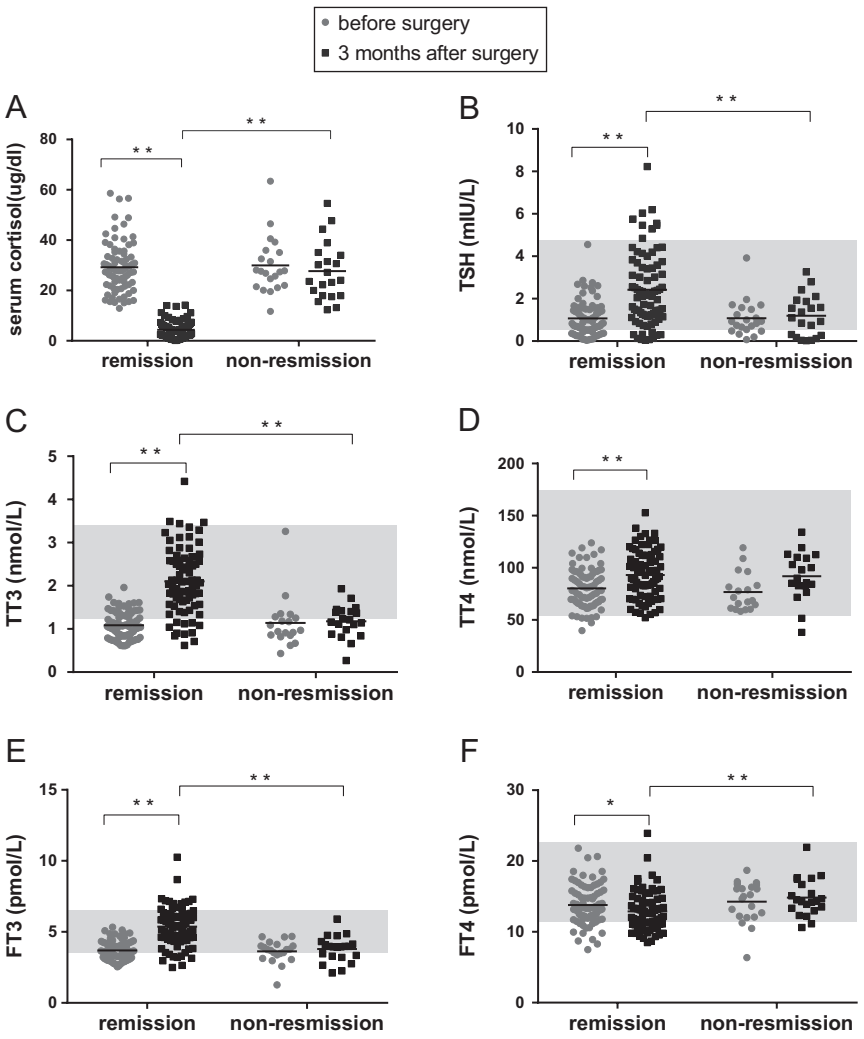

Figure 1
Comparison between remission ($n = 81$) and non-remission ($n = 21$) groups in CD patients before and 3 months after surgery. In the remission group, compared to those before surgery: serum cortisol level (A) dropped significantly; TSH ($P = 0.000$), T3 ($P = 0.000$), T4 ($P = 0.000$) and FT3 ($P = 0.000$) (panel (B, C, D and E)) showed significant increase; FT4 (F) slightly decreased after surgery ($P = 0.012$). Neither serum cortisol nor thyroid hormones showed notable difference before surgery between remission and non-remission group. Three months after surgery, with the reduction of serum cortisol in remission group, TSH (B), T3 (C) and FT3 (E) became significantly lower than those in the non-remission group ($P = 0.000$ for all three indexes). FT4 (F) levels in remission group were significantly lower than those in the non-remission group in the third month postoperative evaluation ($P = 0.004$). The gray bands in panel (B, C, D, E and F) represent the reference range values. The lines represent mean values (±S.D.). *$P < 0.05$, **$P < 0.01$.

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Table 3 Serum cortisol and thyroid functions of 102 CD patients before and 3 months after surgery.

	Remission group (n = 81)			**Non-remission group** (n = 21)				
	Before surgery	3 months after surgery	P^a	Before surgery	3 months after surgery	P^b	P^c	P^d
Serum cortisol (μg/dL)	29.2 ± 9.9	4.4 ± 3.3	0.000	30.0 ± 11.2	27.7 ± 11.6	0.394	0.773	0.000
TSH (mIU/L)	1.07 ± 0.82	2.42 ± 1.73	0.000	1.08 ± 0.84	1.19 ± 0.97	0.443	0.977	0.000
T3 (nmol/L)	1.08 ± 0.31	2.09 ± 0.77	0.000	1.15 ± 0.61	1.16 ± 0.39	0.977	0.535	0.000
T4 (nmol/L)	80.2 ± 18.4	93.1 ± 23.3	0.000	76.6 ± 18.7	91.5 ± 24.5	0.093	0.202	0.668
FT3 (pmol/L)	3.70 ± 0.66	5.37 ± 1.37	0.000	3.63 ± 0.78	3.79 ± 0.91	0.257	0.680	0.000
FT4 (pmol/L)	13.8 ± 2.8	12.9 ± 2.7	0.012	14.3 ± 2.9	14.8 ± 2.7	0.370	0.499	0.004

[a]Comparison between hormones before and 3 months after surgery in remission group. [b]Comparison between hormones before and 3 months after surgery in the non-remission group. [c]Comparison between remission and non-remission group before surgery. [d]Comparison between remission and non-remission group 3 months after surgery.

were not significant, except for FT4's transient decrease on the second month.

In the 28 patients of the remission group undergoing 1-year follow-up (Fig. 3), serum cortisol level markedly dropped after surgery. The median dose of cortisone acetate for post-surgery, 3rd-month, 6th-month and 12th-month replacement treatment were 50.0 mg/day (0~75.0 mg/day), 25.0 mg/day (0~75.0 mg/day), 3.125 mg/day (0~62.5 mg/day) and 0 mg/day (0~25.0 mg/day). The proportion of patients receiving cortisone replacement for post-surgery day, 3rd month, 6th month and 12th month were 85.7, 67.9, 32.1 and 14.3% separately. Compared to those before surgery, TSH, T3 and FT3 showed constant significant increase postoperatively. Elevated TSH above the reference range was found in two, four and three patients on the 3rd, 6th and 12th month. High T3 was seen in only three patients on the third month and then all returned to normal in the 6th- and 12th-month evaluation. Abnormally elevated FT3 was seen in six patients on the 3rd month, two on the 6th month and then only one by 12 months. FT4 level dropped on the 3rd and 6th months but returned to no difference from the baseline level by 12 months. The alternation of T4 was not notable, with 96.4% (27/28) of patients in the remission group constantly maintaining normal T4 level. Overall TPO-Ab and ATG showed no significant change during the 1-year follow-up.

Discussion

The changes of thyroid hormone in hypercortisolemia have been studied for 60 years. The results of previous studies were limited by small sample sizes and incomplete thyroid hormone indexes (8, 9). We investigated thyroid function indexes (including TSH, T3, T4, FT3 and FT4) in 118 CS patients, which, to date, was the largest sample. We found that in the 118 CS patients, the incidence of low TSH, T3 and FT3 was relatively high (TSH, 31%; T3, 69%; FT3, 44%) before treatment. Besides, none of the thyroid function indexes was above the reference range in active CS. In addition, this is also the first report comparing thyroid hormones among CD, adrenal CS and EAS. We found that the levels of FT3 and FT4 in EAS group, who bore the highest serum cortisol level, were lower than those in CD and adrenal CS. Therefore, we recommend

Table 4 Correlations between serum cortisol and thyroid hormones.

			R	P
Before surgery (n = 112)[a]	Serum cortisol	TSH	−0.458[d]	0.000
		T3	−0.358[d]	0.000
		T4	−0.225[c]	0.017
		FT3	−0.449[d]	0.000
		FT4	−0.242[d]	0.009
Three months after surgery (n = 102)[b]	Serum cortisol	TSH	−0.343[d]	0.000
		T3	−0.472[d]	0.000
		T4	−0.076	0.457
		FT3	−0.477[d]	0.000
		FT4	0.216[c]	0.029

[a]Correlations between thyroid hormones and serum cortisol levels were analyzed in all the patients with CS (n = 112) before surgery. [b]Correlations between thyroid hormones and serum cortisol levels were analyzed in all the patients with CD (n = 102) after surgery. [c]$P < 0.05$, [d]$P < 0.01$.

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Table 5 Significant determinants of thyroid hormones in CD (n = 102) with variable parameters in multiple regression analysis.

	Dependent variables	Independent variables	Unstandardized coefficients	Standardized coefficients	*P*	95% CI
			B	Beta		
Before surgery	TSH	F	−0.032	−0.335	0.001	(1.512, 2.675)
	T3	F	−0.011	−0.361	0.000	(−0.017, −0.005)
		BMI	0.021	0.281	0.005	(0.007, 0.035)
	FT3	F	−0.030	−0.447	0.000	(−0.042, −0.018)
		Age	−0.013	−0.225	0.015	(−0.023, −0.003)
		BMI	0.028	0.183	0.047	(0.000, 0.056)
	FT4	Age	−0.058	−0.236	0.025	(−0.109, −0.008)
After surgery	TSH	F	−0.054	−0.356	0.003	(−0.089, −0.020)
	T3	F	−0.034	−0.493	0.000	(−0.050, −0.019)
	FT3	F	−0.065	−0.531	0.000	(−0.090, −0.040)

Parameters analyzed included serum cortisol levels, BMI, gender, age and tumor size (in postoperative analyses, tumor size was excluded).
F: serum cortisol.

testing for CS in patients with low thyroid hormones and low TSH.

In the 102 CD patients, the remission group had higher TSH, T3 and FT3 levels than the non-remission group on the third month after surgery. In the remission group, 11.1% (9/81) TSH, 4.9% (4/81) T3 and 17.3% (14/81) FT3 were above the reference range. Moreover, 14.8% (12/81) of patients in the remission group exhibited elevated thyroid hormones (T3, FT3 and FT4) with nonsuppressed TSH. Within the 3-month monitoring of the nine CD patients in remission group, we observed that TSH increased immediately 1 day after surgery and the increase started to exhibit significance since the second week postoperatively. T3 and FT3 levels went down 1 day after surgery and then rose up to markedly higher levels. Clinicians should be careful not to misdiagnose these conditions as hyperthyroidism and we recommend testing for thyroid function after surgery in patients with CD.

It is well known that glucocorticoids exert an inhibitory action on the hypothalamic–pituitary–thyroid axis (3, 5). Previous reports documented that glucocorticoids decreased the release of TRH and TSH in the hypothalamic and pituitary (11, 12, 13, 14, 15, 16), and increased type 2 deiodinase (D2) activity, which was known to mainly convert T4 to T3 in the central nervous system, and eventually suppressed TSH and thyroid hormone (T3 and T4) secretion (17, 18). The glucocorticoids also decreased type 1 deiodinase (D1), known to convert serum T4 to T3 (17), and that possibly explains the relatively normal T4 and FT4 concentrations in active CS. There have been few studies showing the correlation between serum cortisol and thyroid hormones (19). In our study, we found significant negative correlations between thyroid hormones (TSH, T3 and FT3) and cortisol levels both before and after surgery. In the remission group, as the glucocorticoid concentration went down, TSH, T3 and FT3 increased significantly. TSH's immediate increase after surgery was very likely to be caused by reduced D2 activity in the central nervous system. The mechanism of high T3 and FT3 might be the rise of TSH level and D1 activity in the periphery. A previous study reported elevated FT3 with unsuppressed TSH in CS patients 1 month after surgery (9). They recruited eight patients and measured TSH, FT3 and FT4 only at 1st, 3rd, 6th and 12th month. Our study included closer monitoring of patients' thyroid function postoperatively and we found that TSH increased immediately after surgery. T3 and FT3 went down on the first day after surgery and then rose up notably. The transient decrease of T3 and FT3 may be explained by 'low-T3 syndrome' resulting from surgery stress. Throughout our 1-year follow-up, T4 did not fluctuate as notably as TSH, T3 and FT3 did, and 96.4% (27/28) of patients in the remission group constantly maintained normal T4 level. This may be due to increased conversion from T4 to T3 in the periphery.

Space-occupying lesion of the pituitary is the most common cause of central hypothyroidism, which is defined as low thyroid hormones in conjunction with a low or inappropriately normal TSH level. However, microadenomas are commonly thought unlikely to compromise pituitary functions (20). As is known, microadenomas are quite common in CD. Nestoras Mathioudakis reported (21) that the prevalence of central hypothyroidism was higher in ACTH micros (18%) compared with prolactin micros (1%) and nonfunctioning adenomas (NFAs) (0%). Nevertheless, they found no correlation between free T4 or TSH and the degree of hypercortisolism in that study. In our study, we found no correlation between tumor size and thyroid

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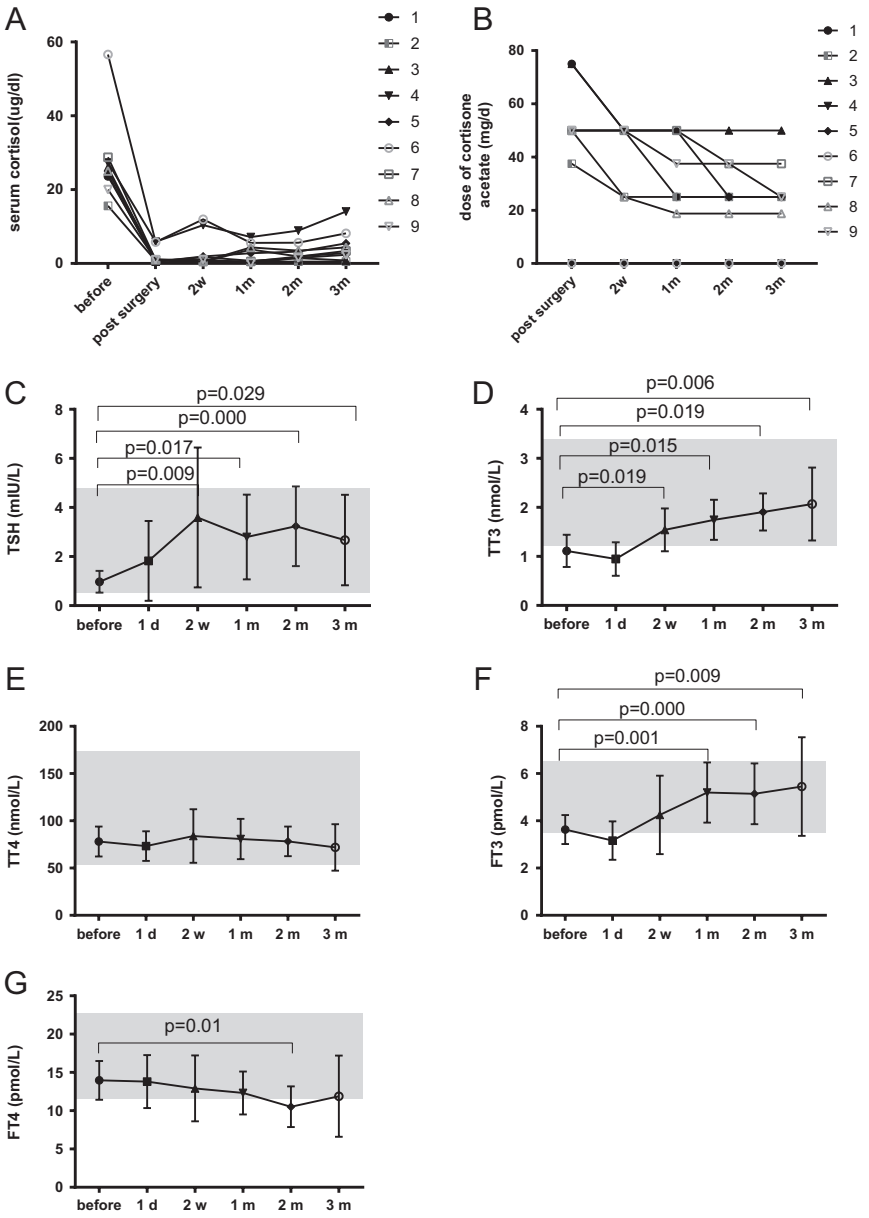

Figure 2
Changes in thyroid function in nine CD patients in the remission group within 3 months after surgery. Serum cortisol (A) markedly dropped after surgery. Panel (B) shows the daily doses of cortisone acetate within 3 months in the nine patients postoperatively. Two of the nine patients received no cortisone replacement after surgery TSH (C), T3 (D), and FT3 (F) showed a significant increase. T4 (E) and FT4 (G) showed no significant change except for the FT4's decrease on the second month. The gray bands in panels C, D, E, F and G represent the reference range values. The lines represent mean values (±S.D.) in panel (C, D, E, F and G).

hormone/serum cortisol. Marked correlations between thyroid hormones and serum cortisol were observed. All patients received transsphenoidal surgery at the same clinic by an experienced surgeon. Therefore, we considered that the changes of thyroid hormones in CD were related to serum cortisol alternation, rather than surgery.

Normalization of hypercortisolism of CS is associated with the induction of autoimmunity (22). Some studies attributed the high TSH with/without low FT4 to exacerbation of underlying autoimmune disease along with the decline of serum cortisol concentration, sometimes accompanied by the increased titer of antithyroid antibodies (23, 24). In our 1-year follow-up (n=28), TSH, T3 and FT3 levels significantly rose up after surgery in the remission group. FT4 levels decreased on the third and sixth months. Moreover, in the hormone evaluation of all the CD patients (n=102), we found that FT4 in the remission group (n=81) became significantly lower after surgery. Also, the postoperative FT4 in the remission group (n=81) was lower than that in the non-remission group (n=21) on the third month. This may be due to transiently increased conversion from T4 to T3 along with the disinhibition of D1. Furthermore, we find no significant fluctuation of anti-thyroperoxidase antibodies or antithyroglobulin antibodies by 1 year after surgery. Therefore, for now, we hold the view that

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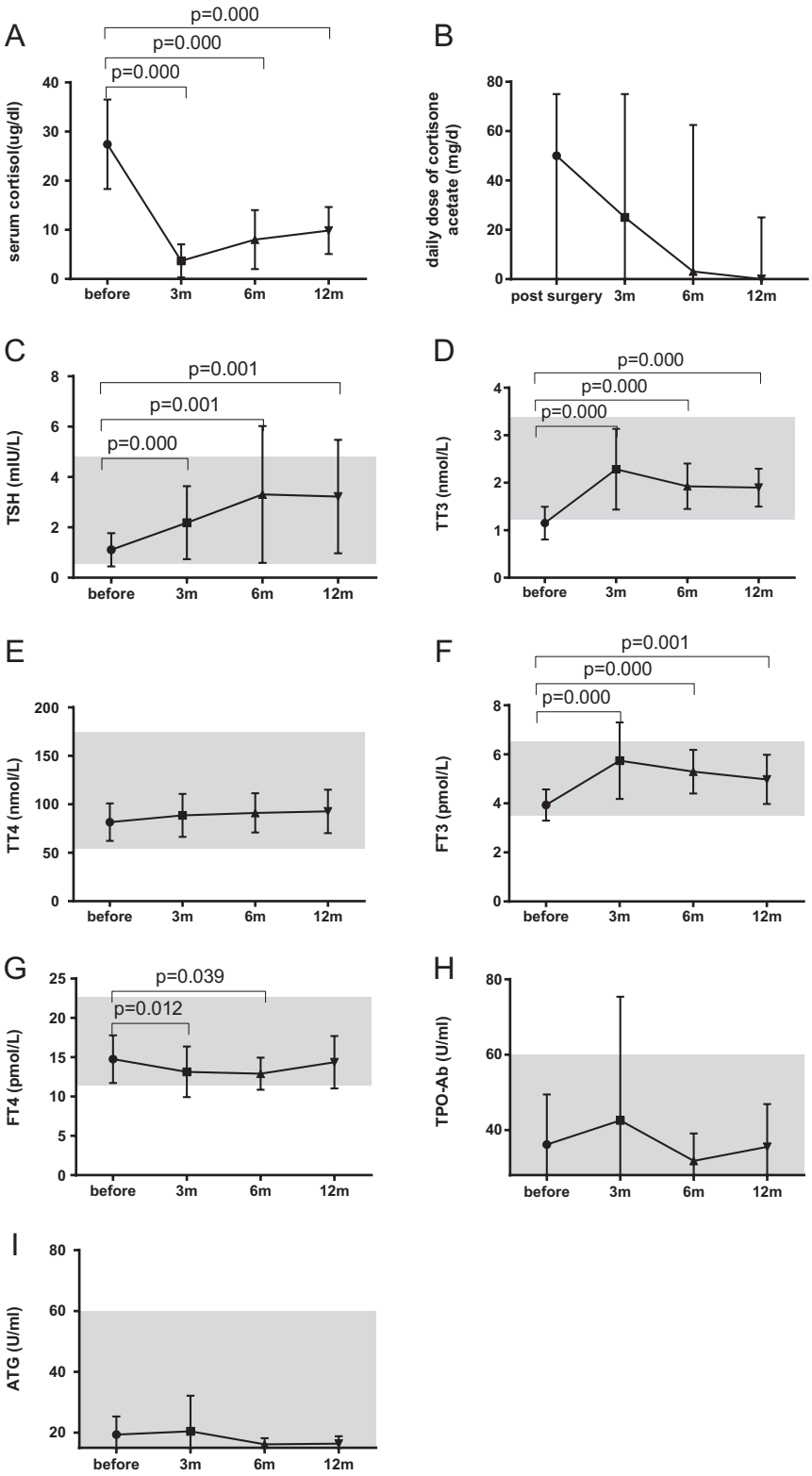

Figure 3
Changes in thyroid hormones, TSH and antithyroid autoantibodies in 28 CD patients in the remission group by 12 months after surgery. Serum cortisol (A) dropped immediately after surgery and then gradually increased. The median dose of cortisone acetate (B) for post-surgery, 3rd-month, 6th-month and 12th-month replacement treatment were 50.0 mg/day, 25.0 mg/day, 3.125 mg/day and 0 mg/day. The proportion of patients receiving cortisone replacement for post-surgery day, 3rd month, 6th month and 12th month were 85.7, 67.9, 32.1 and 14.3% separately. TSH (C), T3 (D) and FT3 (F) showed a significant increase in all the postoperative evaluation. FT4 (G) transiently decreased on the third and sixth month but returned to no difference from the baseline level by 12 months. T4 (E) and antithyroid autoantibodies did not change significantly by 12 months after surgery. The gray bands in panel (C, D, E, F, G, H and I) represent the reference range values. The lines respectively represent median (minimum–maximum) in panel (B), and mean values (±S.D.) in panel (A) and (C, D, E, F, G, H and I).

high TSH with low FT4 after remission of CD is caused by the reduction of serum cortisol, and it should not be defined as primary hypothyroidism. However, as previous studies showed the elevation of thyroid autoantibodies could occur from the second month to the eighth year after surgery (24), further follow-up should be needed for the thyroid autoantibodies to exhibit probable significant alternation.

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https://doi.org/10.1530/EC-19-0309

Limitation

The changes of thyroid functions of adrenal adenoma and EAS after surgery were not elucidated.

Conclusion

Our study revealed the inhibitory action of endogenous hypercortisolemia on hypothalamic–pituitary–thyroid (HPT) axis, in which TSH, T3 and FT3 levels were suppressed. There were negative correlations between serum cortisol and thyroid hormones (TSH, T3 and FT3). After remission of CD, TSH, T3 and FT3 increased significantly, even above the reference range, and mostly returned to normal 1 year after surgery. The changes of thyroid hormones in CD were associated with serum cortisol alternation, not surgery. Antithyroid antibodies showed no significant change after remission of CD. We recommend testing for CS in patients with low thyroid hormones and low TSH. In addition, post-treatment CD patients with high TSH and T3 should be carefully investigated so as not to be misdiagnosed with hyperthyroidism. The molecular mechanism of the changes of thyroid functions after remission of CD is not fully elucidated.

Declaration of interest
The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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Author contribution statement
Hongying Ye designed the research; Boni Xiang, Ran Tao, Xinhua Liu, Min He, Xiaoming Zhu, Yiming Li, Zhenwei Yao, Yongfei Wang, Zengyi Ma, and Hongying Ye diagnosed, treated, followed the patients; Boni Xiang, Ran Tao and Hongying Ye analyzed the data, wrote and edited the manuscript.

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References

1 Nieman LK, Biller BM, Findling JW, Newell-Price J, Savage MO, Stewart PM & Montori VM. The diagnosis of Cushing's syndrome: an Endocrine Society Clinical Practice Guideline. *Journal of Clinical Endocrinology and Metabolism* 2008 **93** 1526–1540. (https://doi.org/10.1210/jc.2008-0125)

2 Lacroix A, Feelders RA, Stratakis CA & Nieman LK. Cushing's syndrome. *Lancet* 2015 **386** 913–927. (https://doi.org/10.1016/S0140-6736(14)61375-1)

3 Fredrickson DS, Forsham PH & Thorn GW. The effect of massive cortisone therapy on measurements of thyroid function. *Journal of Clinical Endocrinology and Metabolism* 1952 **12** 541–553. (https://doi.org/10.1210/jcem-12-5-541)

4 Re RN, Kourides IA, Ridgway EC, Weintraub BD & Maloof F. The effect of glucocorticoid administration on human pituitary secretion of thyrotropin and prolactin. *Journal of Clinical Endocrinology and Metabolism* 1976 **43** 338–346. (https://doi.org/10.1210/jcem-43-2-338)

5 Otsuki M, Dakoda M & Baba S. Influence of glucocorticoids on TRF-induced TSH response in man. *Journal of Clinical Endocrinology and Metabolism* 1973 **36** 95–102. (https://doi.org/10.1210/jcem-36-1-95)

6 Kuku SF, Child DF, Nader S & Fraser TR. Thyrotrophin and prolactin responsiveness to thyrotrophin releasing hormone in Cushing's disease. *Clinical Endocrinology* 1975 **4** 437–442. (https://doi.org/10.1111/j.1365-2265.1975.tb01551.x)

7 Barnett AH, Donald RA & Espiner EA. High concentrations of thyroid-stimulating hormone in untreated glucocorticoid deficiency: indication of primary hypothyroidism? *BMJ* 1982 **285** 172–173. (https://doi.org/10.1136/bmj.285.6336.172-a)

8 Tamada D, Onodera T, Kitamura T, Yamamoto Y, Hayashi Y, Murata Y, Otsuki M & Shimomura I. Hyperthyroidism due to thyroid-stimulating hormone secretion after surgery for Cushing's syndrome: a novel cause of the syndrome of inappropriate secretion of thyroid-stimulating hormone. *Journal of Clinical Endocrinology and Metabolism* 2013 **98** 2656–2662. (https://doi.org/10.1210/jc.2013-2135)

9 Tamada D, Kitamura T, Onodera T, Hamasaki T, Otsuki M & Shimomura I. Clinical significance of fluctuations in thyroid hormones after surgery for Cushing's syndrome. *Endocrine Journal* 2015 **62** 805–810. (https://doi.org/10.1507/endocrj.EJ15-0001)

10 Nieman LK, Biller BM, Findling JW, Murad MH, Newell-Price J, Savage MO, Tabarin A & Endocrine Society. Treatment of Cushing's syndrome: an Endocrine Society clinical practice guideline. *Journal of Clinical Endocrinology and Metabolism* 2015 **100** 2807–2831. (https://doi.org/10.1210/jc.2015-1818)

11 Estupina C, Belmar J, TapiaArancibia L, Astier H & Arancibia S. Rapid and opposite effects of dexamethasone on in vivo and in vitro hypothalamic somatostatin release. *Experimental Brain Research* 1997 **113** 337–342. (https://doi.org/10.1007/bf02450331)

12 Nakagawa K, Ishizuka T, Shimizu C, Ito Y & Wakabayashi I. Increased hypothalamic somatostatin messenger-RNA following dexamethasone administration in rats. *Acta Endocrinologica* 1992 **127** 416–419. (https://doi.org/10.1530/acta.0.1270416)

13 Cintra A, Fuxe K, Wikstrom AC, Visser T & Gustafsson JA. Evidence for thyrotropin-releasing-hormone and glucocorticoid receptor-immunoreactive neurons in various preoptic and hypothalamic nuclei of the male-rat. *Brain Research* 1990 **506** 139–144. (https://doi.org/10.1016/0006-8993(90)91210-8)

14 Kakucska I, Qi YP & Lechan RM. Changes in adrenal status affect hypothalamic thyrotropin-releasing-hormone gene-expression in parallel with corticotropin-releasing hormone. *Endocrinology* 1995 **136** 2795–2802. (https://doi.org/10.1210/endo.136.7.7789304)

15 Perez-Martinez L, Carreon-Rodriguez A, Gonzalez-Alzati ME, Morales C, Charli JL & Joseph-Bravo P. Dexamethasone rapidly regulates TRH mRNA levels in hypothalamic cell cultures: interaction with the cAMP pathway. *Neuroendocrinology* 1998 **68** 345–354. (https://doi.org/10.1159/000054383)

16 Alkemade A, Unmehopa UA, Wiersinga WM, Swaab DF & Fliers E. Glucocorticoids decrease thyrotropin-releasing hormone messenger ribonucleic acid expression in the paraventricular nucleus of the human hypothalamus. *Journal of Clinical Endocrinology and Metabolism* 2005 **90** 323–327. (https://doi.org/10.1210/jc.2004-1430)

17 Chiamolera MI & Wondisford FE. Minireview: thyrotropin-releasing hormone and the thyroid hormone feedback mechanism. *Endocrinology* 2009 **150** 1091–1096. (https://doi.org/10.1210/en.2008-1795)
18 Coppola A, Meli R & Diano S. Inverse shift in circulating corticosterone and leptin levels elevates hypothalamic deiodinase type 2 in fasted rats. *Endocrinology* 2005 **146** 2827–2833. (https://doi.org/10.1210/en.2004-1361)
19 Kitahara H, Imai Y, Yamauchi K, Tomita A & Mizuno S. Pituitary-thyroid function in patients with Cushing's syndrome – comparative study before and after extirpation of adrenal cortex tumor. *Nihon Naibunpi Gakkai Zasshi* 1983 **59** 1086–1098. (https://doi.org/10.1507/endocrine1927.59.8_1086)
20 Freda PU, Beckers AM, Katznelson L, Molitch ME, Montori VM, Post KD, Vance ML & Endocrine Society. Pituitary incidentaloma: an Endocrine Society clinical practice guideline. *Journal of Clinical Endocrinology and Metabolism* 2011 **96** 894–904. (https://doi.org/10.1210/jc.2010-1048)
21 Mathioudakis N, Thapa S, Wand GS & Salvatori R. ACTH-secreting pituitary microadenomas are associated with a higher prevalence of central hypothyroidism compared to other microadenoma types. *Clinical Endocrinology* 2012 **77** 871–876. (https://doi.org/10.1111/j.1365-2265.2012.04442.x)
22 da Mota F, Murray C & Ezzat S. Overt immune dysfunction after Cushing's syndrome remission: a consecutive case series and review of the literature. *Journal of Clinical Endocrinology and Metabolism* 2011 **96** E1670–E1674. (https://doi.org/10.1210/jc.2011-1317)
23 Sahoo JP, Selviambigapathy J, Kamalanathan S, Nagarajan K & Vivekanandan M. Effect of steroid replacement on thyroid function and thyroid autoimmunity in Addison's disease with primary hypothyroidism. *Indian Journal of Endocrinology and Metabolism* 2016 **20** 162–166. (https://doi.org/10.4103/2230-8210.176356)
24 Niepomniszcze H, Pitoia F, Katz SB, Chervin R & Bruno OD. Primary thyroid disorders in endogenous Cushing's syndrome. *European Journal of Endocrinology* 2002 **147** 305–311. (https://doi.org/10.1530/eje.0.1470305)

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