



The management of Cushing's disease — from investigation to treatment

Postępowanie w chorobie Cushinga — od testu diagnostycznego do leczenia

Agata Juszcak, Ashley Grossman

Oxford Centre for Diabetes, Endocrinology and Metabolism, Churchill Hospital, University of Oxford, United Kingdom

Abstract

Cushing's disease (CD) is caused by an adrenocorticotrophin (ACTH) secreting pituitary adenoma and it is the commonest cause of endogenous hypercortisolism. When high suspicion of Cushing's syndrome (CS) exists, recommended screening tests include the overnight dexamethasone suppression test, the low-dose dexamethasone suppression test, or late night salivary cortisol. If the initial test is positive on two occasions, the patient should be referred to a specialist endocrinologist for in-patient assessment, while elevated midnight serum cortisol and a low dose dexamethasone suppression test will confirm endogenous hypercortisolaemia. Plasma ACTH measurement at 9am follows and, if elevated, MRI scan of the pituitary should be performed. Corticotrophin releasing hormone (CRH) test helps to distinguish pituitary from ectopic ACTH-dependent CS, though bilateral petrosal sinus sampling remains the gold standard.

Transsphenoidal surgery is the recognised first-line treatment of CD, and can be repeated if unsuccessful. Second line therapy includes pituitary radiotherapy, bilateral adrenalectomy and medical treatment. Pituitary radiotherapy is very effective but it usually takes several years for its full effect to be seen. Bilateral adrenalectomy is useful in acutely unwell patients, who are unable to tolerate medical therapy. The most effective medical agents inhibit adrenal steroidogenesis and include metyrapone, ketoconazole, mitotane and etomidate. They are used in preparation for surgery, when an operation has been unsuccessful, or when the effects of radiotherapy are being awaited. Cabergoline and pasireotide decrease ACTH production, but are effective in only 30% and 25% of patients, respectively.

It is crucial for patients with CD to be managed in specialist endocrine centres, as the expertise of multidisciplinary team members predicts the best outcome. (*Endokrynol Pol* 2013; 64 (2): 166–174)

Key words: Cushing's disease, investigation, treatment, review

Streszczenie

Choroba Cushinga jest spowodowana przez gruczolaka przysadki, produkującego hormon adrenokortykotropowy (ACTH) i jest najczęstszą przyczyną endogenego hiperkortyzolizmu. W przypadku podejrzenia zespołu Cushinga zalecane testy przesiewowe to test hamowania 1 mg deksametazonu, test hamowania małymi dawkami deksametazonu albo badanie nocnego kortyzolu w ślinie. Jeżeli wynik testu przesiewowego jest 2-krotnie dodatni, pacjent powinien być skierowany do specjalisty–endokrynologa, a stwierdzenie w warunkach szpitalnych podwyższonego stężenia kortyzolu w surowicy o północy i brak hamowania kortyzolu małymi dawkami deksametazonu potwierdza hiperkortyzolemię. Następnie należy oznaczyć stężenie ACTH we krwi o 9. rano i jeżeli jest podwyższone należy wykonać MR przysadki.

Test stymulacji CRH (hormonem uwalniającym hormon kortykotropowy) pomaga odróżnić przysadkowy od ektopowego ACTH-zależnego zespół Cushinga, ale „złotym standardem” pozostaje cewnikowanie zatok skalistych w celu pobrania krwi do oznaczeń ACTH. Operacyjne usunięcie gruczolaka przysadki przez zatokę klinową jest leczeniem pierwszego rzutu w chorobie Cushinga i może być powtórzone jeżeli będzie nieskuteczne. Leczeniem drugiego rzutu są radioterapia przysadki, obustronne usunięcie nadnerczy i leczenie farmakologiczne. Radioterapia przysadki jest bardzo skuteczna, ale pełny efekt leczniczy uzyskujemy po kilku latach. Obustronne usunięcie nadnerczy jest nadal stosowane u ciężko chorych pacjentów, którzy nie tolerują leczenia farmakologicznego. Najbardziej skuteczne leki hamujące syntezę steroidów nadnerczowych to metyrapon, ketokonazol, mitotan i etomidat. Leczenie farmakologiczne stosuje się podczas przygotowania do operacji, kiedy operacja jest nieskuteczna albo w czasie oczekiwania na skutki radioterapii. Kabergolina i pasireotyd obniżają stężenie ACTH, ale są skuteczne odpowiednio tylko w 30% i 25%. Pacjenci z chorobą Cushinga powinni być leczeni w specjalistycznych ośrodkach endokrynologicznych, ponieważ współpraca wielodyscyplinarnego zespołu specjalistów (endokrynologów, neurochirurgów, onkologów i radiologów) decyduje o powodzeniu leczenia. (*Endokrynol Pol* 2013; 64 (2): 166–174)

Słowa kluczowe: choroba Cushinga, badania, leczenie, praca pogładowa

Introduction

Cushing's disease is a rare endocrine disorder, with an incidence of 1–2 cases per million population per year. It results from adrenocorticotrophin (ACTH)-producing pituitary adenomas with a consequential

overproduction of cortisol. It accounts for around 80–85% of patients with ACTH-dependent Cushing's syndrome (CS) and in adults three out of four cases affect women, although in pre-pubertal children it is more common in males [1]. In the 1950s, it was reported that untreated CS had a five-year survival rate of 50%



Prof. Ashley Grossman M.D., Oxford Centre for Diabetes, Endocrinology and Metabolism, Churchill Hospital, Oxford OX3 7LE, UK,
e-mail: ashley.grossman@ocdem.ox.ac.uk

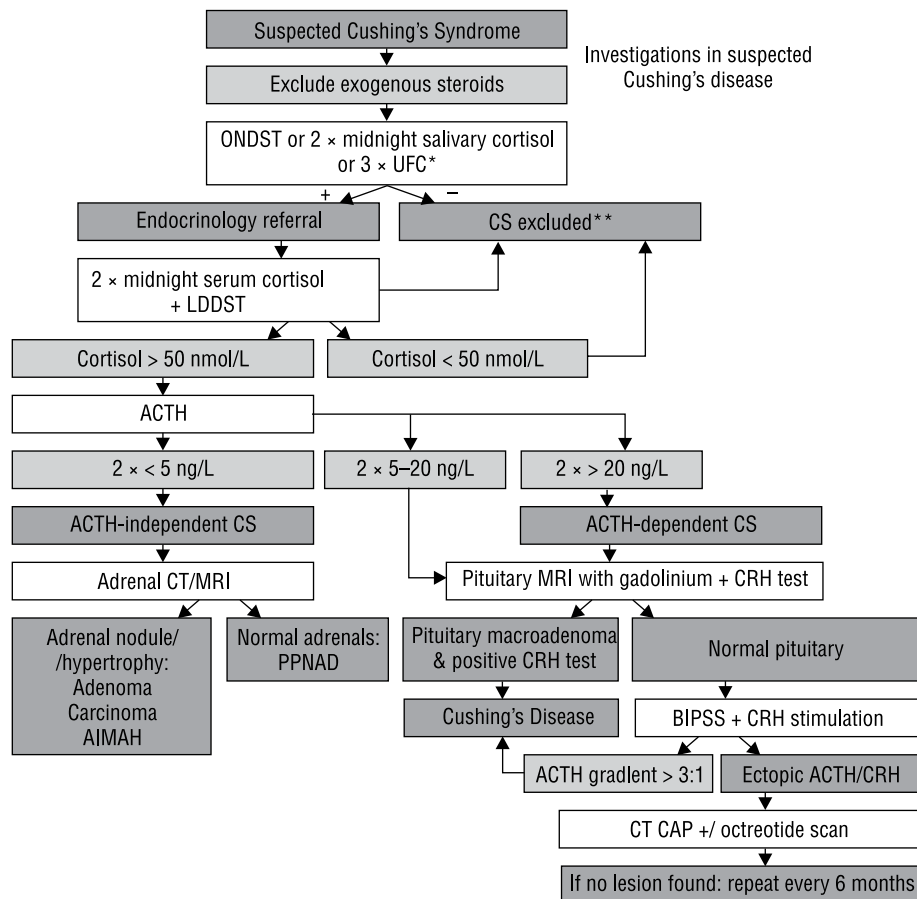


Figure 1. Investigation algorithm in Cushing's syndrome (Adapted with permission from: Juszcak A, Grossman A. The investigation of Cushing syndrome: essentials in optimizing appropriate diagnosis and management. *Ann Saudi Med* 2012; 32: 455–461)

Rycina 1. Algorytm postępowania w zespole Cushinga — optymalne metody diagnostyczne i leczenie (Zaadaptowano z: Juszcak A, Grossman A. The investigation of Cushing syndrome: essentials in optimizing appropriate diagnosis and management. *Ann Saudi Med* 2012; 32: 455–461)

ONDST — overnight dexamethasone suppression test; UFC — urinary free cortisol; CS — Cushing's disease; LDDST — low dose dexamethasone suppression test; ACTH — adrenocorticotrophin hormone; CRH — corticotrophin releasing hormone; AIMAH — ACTH-independent macronodular adrenal hyperplasia; PPNAD — primary pigmented nodular adrenal dysplasia; BIPSS — bilateral inferior petrosal sinus sampling; CT CAP — computed tomography of chest, abdomen and pelvis

due to cardiovascular co-morbidities and infections [2], but recent data suggest survival is similar to an age-matched population when the hypercortisolism has been cured [3] although this has been disputed. If moderate CS persists, the standardised mortality ratio remains increased by between 3.8-fold and 5-fold [4].

The diagnostic pathway for CD starts with a careful history taking and clinical examination. The most discriminatory features of hypercortisolism include purple wide striae, thinned skin with a tendency to bruising, proximal myopathy, facial plethora and osteoporosis at a young age. Other features such as hypertension, central obesity, or impaired glucose tolerance are relatively common and frequently categorised as metabolic syndrome. Acne, hirsutism, dysmenorrhoea and depression overlap with polycystic ovary syndrome and other conditions. In children, new-onset obesity with growth retardation is most characteristic. It is essential to exclude iatrogenic

causes of Cushing's syndrome, as on average 1% of the Western population is using corticosteroids to treat other conditions. It has to be acknowledged that not only oral or injectable preparations may contribute to the development of the features of hypercortisolism, but also topical, inhaled, rectal, intra-articular, nasal and ophthalmic agents.

Investigations (Fig. 1)

When the use of exogenous steroids is excluded, endogenous excess of cortisol has to be confirmed. As a screening test, we recommend one of the following: an overnight (1mg) dexamethasone suppression test or a standard 'low dose' dexamethasone suppression test (LDDST); if available, late night salivary cortisol (two tests) may be useful. We would not routinely recommend 24 hour urinary free cortisol (UFC), but if it is used, a minimum of three collections should be made.

Overnight dexamethasone suppression test (ONDST) and LDDST

The negative feedback regulation of the hypothalamo-pituitary-adrenal axis is used in the investigation of hypercortisolism, as in a healthy individual higher than physiological doses of any glucocorticoid will suppress ACTH and cortisol production.

In the overnight dexamethasone suppression test, 1mg of this steroid is given at midnight and serum cortisol is measured at 9am the next morning. If cortisol suppresses to less than 50 nmol/L, CS is extremely unlikely. The same cut off is used for LDDST, when 0.5 mg of dexamethasone is given every six hours for 48 hours starting at 9 am and cortisol is measured after 48 hours. Both tests have a very high sensitivity of > 95%, but the specificity of the ONDST is 87% or less, while it is in the region of 97–100% for the LDDST [5].

It is important to remember that due to liver metabolism of corticosteroids, all liver enzyme inducers such as anti-epileptics may cause falsely positive results, and liver enzyme inhibitors falsely negative results. CYP 3A4 inducers include phenobarbitone, phenytoin, carbamazepine, rifampicin and pioglitazone, while CYP3A4 inhibitors include cimetidine, itraconazole, diltiazem, ciprofloxacin and fluoxetine [6, 7]. However, all concomitant medication should be suspected. Therefore, if medications affecting corticosteroid metabolism cannot be stopped, different diagnostic tests should be employed.

Assays in current use measure total serum cortisol, and therefore increased cortisol binding globulin in patients taking oestrogens or mitotane, or during pregnancy, will give false positive results. Thus, in such situations a late night salivary cortisol or 24 hour UFC will be of value. If oestrogens in the oral contraceptive pill or as hormone replacement can be stopped, a 4–6 week period is required before the test can be performed.

Late night salivary cortisol and midnight sleeping cortisol

In a healthy individual, serum cortisol reaches its nadir around midnight and starts increasing at 2am–4am, achieving a peak value at 7 am–9 am. This physiological phenomenon is useful in the investigation of Cushing's syndrome, as the circadian rhythmicity of cortisol production is lost in this condition.

Only 3–4% of circulating blood cortisol is unbound, and only this form is excreted in saliva [8]. The level of late night salivary cortisol (SC) correlates well with serum free cortisol, especially if collected using a salivette [9, 10]. Saliva may be collected passively by expectoration as well. A meta-analysis by Carroll et al. showed a pooled sensitivity of 92% and specificity of 96% for diagnosing Cushing's syndrome [11]. Late

night salivary cortisol can be collected at home, it is stable at room temperature, stress-free, and the results are reproducible [12]. Thus, SC appears to be an attractive screening test, especially in children, and when multiple samples are required as in suspected cyclical Cushing's syndrome. It is also recommended in early pregnancy and in women on oral oestrogen preparations, when the increased CBG gives falsely positive results when serum cortisol is measured. However, normative values must be produced for each assay, and it is unclear as to whether the measurement of salivary cortisol is useful in mild CS. Patients should be advised to avoid smoking cigarettes, chewing tobacco or eating liquorice before the test, as these substances block salivary gland 11 β -hydroxysteroid dehydrogenase type 2 (which converts cortisol to cortisone), causing false positive results.

A midnight sleeping serum cortisol is recommended as a confirmatory test for CS, rather than as a screening test, as it requires at least a 48 hour admission to hospital. Ideally, blood should be taken within 15 minutes of waking. Cushing's syndrome can be confidently excluded, even on the basis of a single test, when serum cortisol is less than 50 nmol/L (1.8 μ g/dL) [13]. Some clinicians use a non-sleeping serum midnight cortisol to confirm endogenous hypercortisolism with a cut-off of 207 nmol/L (7.5 μ g/dL), with sensitivity and specificity of more than 96% [14]. However, the specificity decreases to 83% in an obese population [15]. The area between 50 and c.200nmol/L is therefore rather a 'grey' area.

24 hour urinary free cortisol

Unbound cortisol is excreted through the kidneys and most of it is reabsorbed. Therefore only a small fraction can be measured in urine. The measurement of 24 hour urinary free cortisol for the diagnosis of Cushing's syndrome has a high sensitivity of 96% if a low threshold is used, but its specificity is then poor at 40–50% [13]. Therefore, we generally discourage its use as a screening test. If it is done, at least three tests are needed. In the study by Nieman et al., even in subsequently histologically-proven Cushing's disease, one out of four 24 hour UFCs was normal in 11% of patients [16]. This test cannot be relied upon in patients with renal impairment or with fluid intake of more than 5L/24 hours, as in the latter it results in a false positive result [17]. There are interactions with carbenoxolone, carbamazepine, finofibrate and liquorice [6].

Further investigations

When there is a high clinical suspicion of Cushing's syndrome, and two of the screening tests are positive, the patient should be referred to a specialist endocri-

nologist for further investigations and treatment. In our centre, the patient would be admitted to hospital and midnight sleeping serum cortisol would be checked twice, followed by the LDDST with a baseline ACTH level. If cortisol is not suppressed below 50 nmol/L, the CRH (corticotrophin-releasing hormone) stimulation test is arranged, with measurement of both ACTH and cortisol.

One should remember that in cases of high clinical suspicion of the diagnosis and negative tests, cyclical Cushing's syndrome should be excluded; therefore, tests should be repeated on several occasions and particularly at a time when the disease is clinically active. The prevalence of cyclical Cushing's syndrome is reported as 15% and is more common in women [18].

Plasma ACTH and the CRH stimulation test

As soon as hypercortisolism is confirmed, plasma ACTH at 9 am should be checked as this will guide the differential diagnosis. A blood sample for an ACTH needs to be spun immediately and the plasma frozen, otherwise results received may be falsely low. An ACTH above 15–20 ng/L suggests ACTH-dependent CS, and less than 10 ng/L suggests ACTH-independence. For results falling between these values, the CRH test should be used to differentiate between Cushing's disease and ectopic CS from primary adrenal pathology [1].

There are some hints which point towards ectopic ACTH-dependent CS: often, the hypercortisolaemic features are rapid in onset (a few weeks to a few months), and hypokalaemia occurs in almost 100% of patients and only in 10% of patients with CD (possibly more frequently in patients with macroadenomas [19]). This is a consequence of higher levels of cortisol and saturation of 11 β -hydroxysteroid dehydrogenase type 2, which is responsible for converting cortisol to cortisone and preventing its mineralocorticoid action.

Generally, random plasma ACTH and serum cortisol levels in patients with ectopic CS are higher than in CD, and on the LDDST patients with CD can usually suppress their cortisol by 30%, while the response is flat in patients with ectopic CS [20]. An exception to these general rules occurs in patients with CD secondary to a pituitary macroadenoma, which comprise 5–10% of CD cases and may behave biochemically like ectopic CS.

The CRH test helps to differentiate ACTH-dependent CS further. In adults, 100 μ g of human or ovine sequence corticotrophin-releasing hormone is injected intravenously and ACTH and cortisol responses measured at baseline and at 15, 30, 45, 60 and 90 minutes. In CD, ACTH tends to increase by 35–50% and cortisol by at least 20%, which is not the case in an ectopic ACTH-dependent CS [21]. Such results are explained by the frequent presence of CRH1 receptors on the cells of

pituitary corticotroph adenomas, although some 7–14% of patients with subsequent histologically-proven CD do not show a response [13].

HDDST

Previously, the high-dose dexamethasone suppression test followed the LDDST, when 2 mg of dexamethasone was given every six hours for 48 hours and cortisol was measured six hours after the last dose. It was reported that around 80% of patients with CD suppressed cortisol by 50% from baseline, which was not observed in ectopic CS [13]. However, in many endocrine centres, where inferior petrosal sinus sampling is available, this test has been abandoned as it provided little additional diagnostic information, and is relatively risky.

Imaging

Pituitary

An MRI scan of the pituitary gland is the preferred imaging mode in CD. An unenhanced scan will visualise a pituitary adenoma in 50% of patients, which increases only to 50–60% on images with gadolinium contrast. In the majority of cases, a microadenoma is found with mean diameter of 6mm, which appears hypodense on pre-contrast imaging and is not enhanced after intravenous gadolinium in 95%, while in 5% it may become isodense [22, 23]. Dynamic contrast imaging may make identification more accurate. In 5–10% of patients with CD, a pituitary macroadenoma is present. It should also be acknowledged that in 10% of a population having brain imaging for different reasons, a pituitary 'incidentaloma' may be visualised [24].

Adrenals

There is no particular need to image the adrenal glands if biochemical investigations point towards Cushing's disease. In patients who did have their adrenals scanned with CT, bilateral adrenal hyperplasia has been found in the majority (70%) of patients with CD and in all patients with an ectopic source [25].

Bilateral inferior petrosal sinus sampling (BIPSS)

BIPSS is the most accurate test for diagnosing Cushing's disease, with a sensitivity of up to 97% and specificity of up to 100% when CRH stimulation is used [26]. However, its success rate depends on the experience of the radiologist and the number of procedures performed per year. In our endocrine centre, BIPSS is performed for almost all patients with ACTH-dependent Cushing's syndrome, except for patients with pituitary macroadenomas. The catheterisation of IPS is confirmed by venography and, if this fails, a sample is taken from a high internal jugular vein. We use 100 μ g of human CRH

given i.v. for stimulation of ACTH to enhance specificity; a 10 μg injection of desmopressin can be used as an alternative, but the results for desmopressin are currently based on small numbers of patients. ACTH is sampled simultaneously from bilateral IPS and peripheral blood at 0, 3–5, 8–10 and 12–15 minutes. CD is biochemically proven if the IPS-to-peripheral ACTH ratio exceeds 2:1 on basal sampling or 3:1 after stimulation with CRH; in ectopic ACTH-dependent CS the ratio remains below 2:1 (regardless of stimulation) with a mean value of 1.3 ± 0.1 [26]. In the St. Bartholomew's Hospital cohort, in the majority of patients a diagnostic IPS/peripheral ratio was achieved on 5 minutes sampling, so we suggest that 12–15 minutes ACTH measurement is not necessary.

A successful stimulated BIPSS can be used for pre-operative lateralisation of the adenoma with a ratio above 1:1.4 between inferior petrosal sinuses being significant [26, 27], although some centres find this unreliable. It has been reported concordant with the surgical findings in 72–83% of adult patients, with slightly higher rates in children [28].

Treatment

The aim of the treatment of CD is to restore normocortisolaemia and improve, or if possible reverse, its consequences. This can be achieved by transsphenoidal surgery, radiotherapy, bilateral adrenalectomy or medical treatment. In cases of CD caused by a macroadenoma, a pressure effect on the optic chiasm or third nerve may need to be relieved urgently by transsphenoidal surgery.

Transsphenoidal surgery

Transsphenoidal surgery (TSS) is regarded as a first-line treatment for CD. Ideally, a micro-adenectomy should be performed, but if the tumour is not visible during surgery, hemiphysectomy could be an option, guided by IPSS results and MRI findings. If a macroadenoma with suprasellar extension is identified on pre-operative imaging, an operation is still indicated but may not be curative.

Biochemically, the aim of surgery for CD is a 9 am serum cortisol during the first week after surgery of less than 50 nmol/L (1.8 $\mu\text{g}/\text{L}$), after withholding hydrocortisone replacement for at least 12 hours, although remission with a serum cortisol between 50 and 300 nmol/L (1.8–10.8 $\mu\text{g}/\text{L}$) on a five-point cortisol day-curve is still compatible with success. Persistent disease is defined as a serum cortisol of more than 300 nmol/L [29]. The reported remission rate for microadenomas ranges between 65% and 90% [29–31], while for macroadenomas it is only 35–60%. Recurrence is most common in the first two years after operation [32]. Recurrence has been

reported in 5–10% of patients five years after surgery and in 15–20% after ten years [3, 33], and can occur even in patients considered 'cured' by the most stringent criteria. The best predictor of long-term cure seems to be lack of recovery of the hypothalamo-pituitary-adrenal axis in the first three years after operation [33].

If hypercortisolism persists despite surgery, then re-operation is possible especially if total hypophysectomy is thought to be suitable; however, remission rates are lower and range between 50% and 70% in microadenomas [34]. In some centres, TSS is repeated in the first week post the initial operation if a cure is not achieved, but delayed control of hypercortisolism over 4–6 weeks has been reported in some cases [35].

Radiotherapy

External beam radiotherapy (RT) is considered a second line treatment for persistent or recurrent CD, if repeated surgery is not recommended. Currently, all radiotherapy is stereotactic and is delivered as conventional RT in 25–30 fractions of total dose of 45–50 Gy, or as highly-focused radiosurgery in a single session of 18–24 Gy. For radiosurgery, the pituitary adenoma needs to be at least 5 mm away from the optic chiasm to lower the risk of its injury [36]. Hypopituitarism has been reported in 20–40% of patients over ten years following RT, and optic neuropathy in 0–4% [37].

While RT can control hypercortisolism, there is often a considerable delay in its onset of effectiveness; therefore, medical treatment is required in the meantime. Tumour growth control has been reported to be achieved in 93–100% of patients at eight years, and biochemical remission in 46–74% of cases with fractionated conventional RT [38]. In the published literature, there is no direct comparison of the effects of conventional RT versus 'gamma-knife' radiosurgery. Radiosurgery has been reported to control CD biochemically in 42–54% of patients at 45–55 months from its delivery, with the onset of remission at 13–22 months [39]. The onset of eucortisolism appears to be more rapid in children, often occurring in less than 12 months.

Bilateral adrenalectomy

Bilateral adrenalectomy still remains a valuable option in the treatment of severe Cushing's disease in acutely unwell patients, when control of hypercortisolism is required immediately and the patient cannot tolerate medical therapy. It is also recommended when previous transsphenoidal surgery, radiotherapy and medical agents have failed to control the features of CD. It may be the treatment of choice following unsuccessful pituitary surgery for women, who seek fertility and do not wish to risk gonadotrophin deficiency secondary to radiotherapy [29]. This

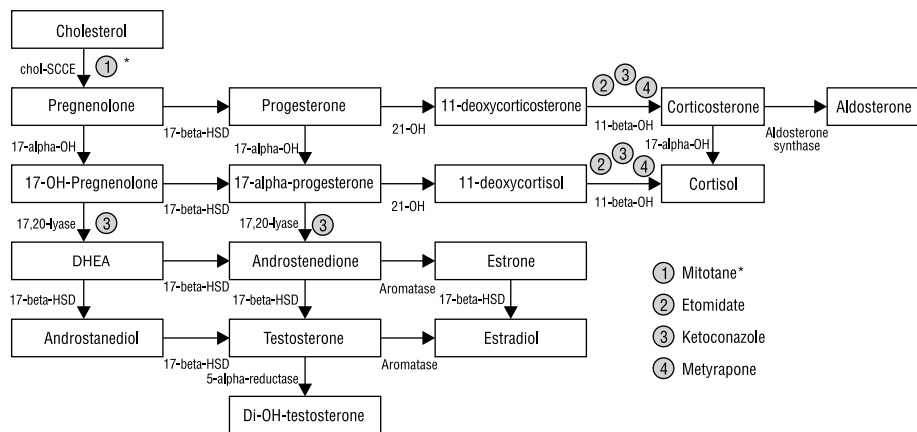


Figure 2. Adrenal steroidogenesis

Rycina 2. Steroidogeneza w nadnerczach

*Mitotane is an adrenolytic agent, the mechanism of action of which remains not fully understood

DHEA — dihydroepiandrosterone; Enzymes: chol-SCCE — cholesterol side-chain cleavage enzyme, 17-alpha-OH — 17-alpha-hydroxylase, 17-beta-HSD — 17-beta-hydroxysteroid dehydrogenase, 21-OH — 21-hydroxylase, 11-beta-OH — 11-beta-hydroxylase

procedure ideally should be done laparoscopically and requires subsequent lifelong replacement of gluco- and mineralocorticoids.

Bilateral adrenalectomy is a definitive treatment of hypercortisolism except for metastatic adrenal carcinoma, in which case cortisol levels may be decreased only temporarily. It does not reduce ACTH levels, and therefore hyperpigmentation may continue to worsen and surveillance with annual MRI scan of the pituitary gland and ACTH is required to detect Nelson's syndrome (NS). NS is caused by autonomous growth of a corticotroph adenoma following bilateral adrenalectomy for CD and is reported in 30-50% of patients. Neoadjuvant radiotherapy is used in some centres and has been proven to prevent it in up to 50% of cases [40, 41]. However, with modern MRI available, we usually prefer to delay radiotherapy unless and until there is clear evidence of tumour progression.

Medical treatment

The medical treatment of CD is used to control hypercortisolism, when transsphenoidal surgery is awaited, has been unsuccessful, or when the effects of radiotherapy are expected but delayed. The main agents used are adrenal steroidogenesis inhibitors: metyrapone, ketoconazole, mitotane, and etomidate; these remain the most effective. They do not affect ACTH levels or tumour growth but reduce cortisol production and improve the features of CS such as hyperglycaemia, hypertension or fluid retention. If steroidogenesis inhibitors are poorly tolerated or CD is mild, centrally-acting agents may be used, such as cabergoline or pasireotide, though their efficiency is less spectacular. All the above agents aim for a mean serum cortisol of

150–300 nmol/L on a three-point or five-point cortisol day curve. A different mechanism of action applies to mifepristone, which is a glucocorticoid receptor antagonist, but experience with this medication is limited.

Metyrapone

Metyrapone inhibits 11 β -hydroxylase (Fig. 2), and therefore the production of cortisol and aldosterone. This results in an excess of 11-deoxycorticosterone, which can cross-react in cortisol assays [42], increased 11-deoxycorticosterone, which has mineralocorticoid properties, and excess of adrenal androgens.

In our endocrine unit, it is the first line medical agent used in the treatment of CD, but we are aware that its availability may be limited in some countries. The starting dose of metyrapone is 250–500 mg three times a day orally, with a maximal dose of 1.5 g four times a day. It significantly decreases cortisol level after two hours and mean cortisol level below 400 nmol/L is usually achieved after a median time of two weeks in the majority of patients [43]. Most of its side effects overlap with features of hypercortisolism and include nausea and vomiting, worsening hypokalaemia, hypertension, oedema, hirsutism and acne. Therefore, long-term use is not advised in young women, where ketoconazole would be preferred [29].

Ketoconazole

Ketoconazole is an anti-fungal agent which inhibits 11 β -hydroxylase and 17, 20-lyase in the steroid synthetic pathway (Fig. 2) and it decreases the production of cortisol and androgens. The recommended initial dose is 200 mg twice a day, which if tolerated could be increased to 400 mg three times a day as a maximal

dose. It takes several weeks to achieve its full effect on hypercortisolism, and with careful cortisol monitoring, hypoadrenalism is very rarely seen. It has to be remembered that proton pump inhibitors should be discontinued while on this treatment, as gastric acidity is required for drug absorption. Long-term treatment with ketoconazole has been reported to control hypercortisolism in 51.5% of patients [44].

The commonest adverse effects reported include gastro-intestinal upset, rash or deranged liver enzymes (elevated aminotransferases and GGT), which could be asymptomatic or cause right upper quadrant pain with acute liver failure in one in 15,000 cases [45]. Up to three times the upper normal range elevation in liver enzymes levels does not contraindicate treatment with ketoconazole, but liver function needs to be carefully monitored [29]. Usually, a rash or hepatotoxicity will present in the first week of treatment or after increasing the dose [42]. Men may report deterioration of libido, erectile dysfunction or gynaecomastia due to its anti-androgenic effect, so morning testosterone levels may need to be monitored.

Mitotane

Mitotane is an oral adrenolytic agent; it is sometimes used as a third line of medical treatment for severe CD, but most of the experience with this drug comes from the treatment of adrenal adenocarcinoma secreting cortisol. The initial recommended dose is 500 mg at night, increased gradually to 1 g three times a day, if tolerated. Its onset of action is delayed to 4–6 weeks and it has got a long half-life, which is related to its accumulation in fat tissue. Therefore, even after stopping mitotane, its effects persist for several weeks or even months [46]. Mitotane elevates the level of CBG, so measuring serum cortisol is not very helpful in monitoring the treatment. The serum level of mitotane should be checked to avoid toxic levels and UFC may be of some assistance. If a 'block-and-replace' regimen is used, it has to be acknowledged that mitotane increases the metabolism of exogenous steroids and so the dose prescribed ought to be about 30% higher [47].

Mitotane treatment is usually limited by several side effects, which are almost invariable at higher doses. Among those reported are gastro-intestinal upset, anorexia, ataxia, vertigo, abnormal liver and thyroid function, hyperuricaemia and hypercholesterolaemia [42, 45].

Etomidate

Etomidate is an anaesthetic agent and the only intravenous preparation available in the treatment of acutely unwell patients with CD who are unable to take oral medications and are unfit for surgery. It blocks 11 β - and 18-hydroxylase as well as 17, 20-lyase (Fig. 2).

Etomidate controls hypercortisolism over the first 12 hours of treatment [48] and may cause hypoadrenalism, and therefore levels of cortisol and potassium need to be monitored every four hours. For that reason, and because it is a sedating agent, it should generally be administered in an intensive care setting. A starting dose of 2 mg/h has been reported, and is used in our centre, and the dose should be titrated according to cortisol levels, aiming for a serum cortisol of 500–800 nmol/L in acutely unwell patients and 150–300 nmol/L when the acute illness has been resolved. A simultaneous infusion of hydrocortisone of 0.5–2 mg/h may be required [49].

Cabergoline

Dopamine receptor agonists, such as bromocriptine or cabergoline, work centrally and data are limited regarding their efficacy in Cushing's disease. D2 receptors have been found on the cells of corticotroph adenomas in 75–83% of cases [50]. Single case series reported variable responses to bromocriptine from 1–50% measured as a decrease in ACTH and cortisol, and tumour shrinkage in 20% of subjects. However, the effect is weak [51]. Cabergoline at a dose of 1–7 mg weekly has been reported to control hypercortisolaemia of CD in 25–40% of patients in small series [50, 52, 53]. In a recent study of 30 patients by Godbout et al., cabergoline at a mean dose of 2 mg/week controlled UFC levels and the clinical features of CD in 30% of patients with persistent or recurrent CD over a mean period of 37 months [52]. Dopamine receptor agonists are usually well tolerated and common side effects include nausea and dizziness. Cardiac valvulopathy related to this treatment remains anecdotal at the doses used for the treatment of pituitary tumours, and should not be a cause for concern.

Pasireotide

Pasireotide (SOM230, Novartis) is a novel somatostatin analogue with affinity to most of somatostatin receptors: sst1-3 and sst5. It looked promising as according to one study 38% of corticotroph adenoma cells express sst5 [54]. Colao et al. published the results of a phase III multicentre trial involving 162 patients with CD, who received 600 μ g or 900 μ g of pasireotide via a twice a day subcutaneous injection. After 12 months of treatment, only 25% on the higher dose and 13% on the lower dose normalised UFC levels and it was noted that mainly patients with mild to moderate hypercortisolaemia responded [55, 56]; 73% of participants experienced new or worsening hyperglycaemia despite decreasing cortisol levels, and 46% required anti-glycaemic medications to control it. Mild transient elevation of liver enzymes was noted in 29% of patients, diarrhoea and nausea occurred in more than 50% of cases with cholelithiasis in 30%.

Mifepristone

Mifepristone is a progesterone and glucocorticoid receptor antagonist, better known as an agent used in medical abortion. Experience with this medication is limited in CD and it should be considered only as an adjuvant therapy. The levels of ACTH and cortisol remain elevated on this treatment, and therefore monitoring is based on clinical assessment. High levels of cortisol act on the mineralocorticoid receptor and so hypertension, water retention and hypokalaemia are significant problems. Following the study by Flesariu et al. on 43 patients with CD treated with this agent [57], it was approved by the FDA for treatment of hyperglycaemia related to CS, as it showed improvement in area under the curve for glucose in GTT in patients with pre-diabetes and improvement of HbA1c from 7.4% to 6.2% with pre-existing diabetes. The study included ten patients with pituitary macroadenoma, of which one was reported to have tumour enlargement at the six month follow-up scan.

Follow-up

Even patients with cured CD require life-long follow-up, as even 20 years after successful surgery, relapse may occur. In our centre, patients in remission/cured are assessed every year [33].

Conclusions

Cushing's disease, despite being a rare endocrine disorder, is associated with significant morbidities. It should be investigated and managed in an endocrine centre with access to appropriate tests and procedures. Therefore, it is best managed by a multidisciplinary team with an experienced neurosurgeon, neuroradiologist and oncologist. The treatment of Cushing's disease may prove challenging, but the achievement of a long-term cure transforms patients' quality of life and can be extremely gratifying for the treating physician.

References

- Newell-Price J, Bertagna X, Grossman AB et al. Cushing's syndrome. *Lancet* 2006; 367: 1605–1617.
- Plotz CM, Knowlton AI, Ragan C. The natural history of Cushing's syndrome. *Am J Med* 1952; 13: 597–614.
- Swearingen B, Biller BM, Barker FG, 2nd et al. Long-term mortality after transsphenoidal surgery for Cushing disease. *Ann Intern Med* 1999; 130: 821–824.
- Lindholm J, Juul S, Jorgensen JO, et al. Incidence and late prognosis of Cushing's syndrome: a population-based study. *J Clin Endocrinol Metab* 2001; 86: 117–123.
- Kennedy L, Atkinson AB, Johnston H et al. Serum cortisol concentrations during low dose dexamethasone suppression test to screen for Cushing's syndrome. *Br Med J (Clin Res Ed)* 1984; 289: 1188–1191.
- Nieman LK, Biller BM, Findling JW, et al. The diagnosis of Cushing's syndrome: an Endocrine Society Clinical Practice Guideline. *J Clin Endocrinol Metab* 2008; 93: 1526–1540.
- Putignano P, Kaltsas GA, Satta MA et al. The effects of anti-convulsant drugs on adrenal function. *Horm Metab Res* 1998; 30: 389–397.

- Raff H, Raff JL, Findling JW. Late-night salivary cortisol as a screening test for Cushing's syndrome. *J Clin Endocrinol Metab* 1998; 83: 2681–2686.
- Kahn JP, Rubinow DR, Davis CL et al. Salivary cortisol: a practical method for evaluation of adrenal function. *Biol Psychiatry* 1988; 23: 335–349.
- Poll EM, Kreitschmann-Andermahr J, Langejuergen Y et al. Saliva collection method affects predictability of serum cortisol. *Clin Chim Acta* 2007; 382: 15–19.
- Carroll T, Raff H, Findling JW. Late-night salivary cortisol for the diagnosis of Cushing syndrome: a meta-analysis. *Endocr Pract* 2009; 15: 335–342.
- Viardot A, Huber P, Puder JJ et al. Reproducibility of nighttime salivary cortisol and its use in the diagnosis of hypercortisolism compared with urinary free cortisol and overnight dexamethasone suppression test. *J Clin Endocrinol Metab* 2005; 90: 5730–5736.
- Newell-Price J, Trainer P, Besser M et al. The diagnosis and differential diagnosis of Cushing's syndrome and pseudo-Cushing's states. *Endocr Rev* 1998; 19: 647–672.
- Papanicolaou DA, Yanovski JA, Cutler GB et al. A single midnight serum cortisol measurement distinguishes Cushing's syndrome from pseudo-Cushing states. *J Clin Endocrinol Metab* 1998; 83: 1163–1167.
- Putignano P, Bertolini M, Losa M et al. Screening for Cushing's syndrome in obese women with and without polycystic ovary syndrome. *J Endocrinol Invest* 2003; 26: 539–544.
- Newell-Price J, Jorgensen JO, Grossman A. The diagnosis and differential diagnosis of Cushing's syndrome. *Horm Res* 1999; 51 (Suppl. 3): 81–94.
- Mericq MV, Cutler GB, Jr. High fluid intake increases urine free cortisol excretion in normal subjects. *J Clin Endocrinol Metab* 1998; 83: 682–684.
- Alexandraki KI, Kaltsas GA, Isidori AM et al. The prevalence and characteristic features of cyclicality and variability in Cushing's disease. *Eur J Endocrinol* 2009; 160: 1011–1018.
- Newell-Price J, Grossman AB. Differential diagnosis of Cushing's syndrome. *Arq Bras Endocrinol Metabol* 2007; 51: 1199–1206.
- Isidori AM, Kaltsas GA, Mohammed S et al. Discriminatory value of the low-dose dexamethasone suppression test in establishing the diagnosis and differential diagnosis of Cushing's syndrome. *J Clin Endocrinol Metab* 2003; 88: 5299–5306.
- Newell-Price J, Morris DG, Drake WM et al. Optimal response criteria for the human CRH test in the differential diagnosis of ACTH-dependent Cushing's syndrome. *J Clin Endocrinol Metab* 2002; 87: 1640–1645.
- Buchfelder M, Nistor R, Fahlbusch R et al. The accuracy of CT and MR evaluation of the sella turcica for detection of adrenocorticotrophic hormone-secreting adenomas in Cushing disease. *AJNR Am J Neuroradiol* 1993; 14: 1183–1190.
- Findling JW, Doppman JL. Biochemical and radiologic diagnosis of Cushing's syndrome. *Endocrinol Metab Clin North Am* 1994; 23: 511–537.
- Buurman H, Saeger W. Subclinical adenomas in postmortem pituitaries: classification and correlations to clinical data. *Eur J Endocrinol* 2006; 154: 753–758.
- Sahdev A, Reznick RH, Evanson J et al. Imaging in Cushing's syndrome. *Arq Bras Endocrinol Metabol* 2007; 51: 1319–1328.
- Kaltsas GA, Giannulis MG, Newell-Price JD et al. A critical analysis of the value of simultaneous inferior petrosal sinus sampling in Cushing's disease and the occult ectopic adrenocorticotropin syndrome. *J Clin Endocrinol Metab* 1999; 84: 487–492.
- Oldfield EH, Chrousos GP, Schulte HM et al. Preoperative lateralization of ACTH-secreting pituitary microadenomas by bilateral and simultaneous inferior petrosal venous sinus sampling. *N Engl J Med* 1985; 312: 100–103.
- Lienhardt A, Grossman AB, Dacie JE et al. Relative contributions of inferior petrosal sinus sampling and pituitary imaging in the investigation of children and adolescents with ACTH-dependent Cushing's syndrome. *J Clin Endocrinol Metab* 2001; 86: 5711–5714.
- Biller BM, Grossman AB, Stewart PM et al. Treatment of adrenocorticotropin-dependent Cushing's syndrome: a consensus statement. *J Clin Endocrinol Metab* 2008; 93: 2454–2462.
- Hassan-Smith ZK, Sherlock M, Reulen RC et al. Outcome of Cushing's disease following transsphenoidal surgery in a single center over 20 years. *J Clin Endocrinol Metab* 2012; 97: 1194–1201.
- Hofmann BM, Fahlbusch R. Treatment of Cushing's disease: a retrospective clinical study of the latest 100 cases. *Front Horm Res* 2006; 34: 158–184.
- Storr HL, Alexandraki KI, Martin L et al. Comparisons in the epidemiology, diagnostic features and cure rate by transsphenoidal surgery between paediatric and adult-onset Cushing's disease. *Eur J Endocrinol* 2011; 164: 667–674.
- Alexandraki KI, Kaltsas G, Isidori AM et al. Long-Term Remission and Recurrence Rates in Cushing's Disease: Predictive Factors in a Single-Centre Study. *Eur J Endocrinol* 2013; 168: 639–648.
- Hofmann BM, Hlavac M, Kreutzer J et al. Surgical treatment of recurrent Cushing's disease. *Neurosurgery* 2006; 58: 1108–1118.
- Valassi E, Biller BM, Swearingen B et al. Delayed remission after transsphenoidal surgery in patients with Cushing's disease. *J Clin Endocrinol Metab* 2010; 95: 601–610.

36. Leber KA, Bergloff J, Pendl G. Dose-response tolerance of the visual pathways and cranial nerves of the cavernous sinus to stereotactic radiosurgery. *J Neurosurg* 1998; 88: 43–50.
37. Minniti G, Brada M. Radiotherapy and radiosurgery for Cushing's disease. *Arq Bras Endocrinol Metabol* 2007; 51: 1373–1380.
38. Minniti G, Osti M, Jaffrain-Rea ML et al. Long-term follow-up results of postoperative radiation therapy for Cushing's disease. *J Neurooncol* 2007; 84: 79–84.
39. Castinetti F, Nagai M, Dufour H et al. Gamma knife radiosurgery is a successful adjunctive treatment in Cushing's disease. *Eur J Endocrinol* 2007; 156: 91–98.
40. Jenkins PJ, Trainer PJ, Plowman PN et al. The long-term outcome after adrenalectomy and prophylactic pituitary radiotherapy in adrenocorticotropin-dependent Cushing's syndrome. *J Clin Endocrinol Metab* 1995; 80: 165–171.
41. Gil-Cardenas A, Herrera MF, Diaz-Polanco A et al. Nelson's syndrome after bilateral adrenalectomy for Cushing's disease. *Surgery* 2007; 141: 147–151.
42. Dang CN, Trainer P. Pharmacological management of Cushing's syndrome: an update. *Arq Bras Endocrinol Metabol* 2007; 51: 1339–1348.
43. Verhelst JA, Trainer PJ, Howlett TA et al. Short and long-term responses to metyrapone in the medical management of 91 patients with Cushing's syndrome. *Clin Endocrinol (Oxf)* 1991; 35: 169–178.
44. Castinetti F, Morange I, Jaquet P et al. Ketoconazole revisited: a preoperative or postoperative treatment in Cushing's disease. *Eur J Endocrinol* 2008; 158: 91–99.
45. Praw SS, Heaney AP. Medical treatment of Cushing's disease: Overview and recent findings. *Int J Gen Med* 2009; 2: 209–217.
46. Luton JP, Cerdas S, Billaud L et al. Clinical features of adrenocortical carcinoma, prognostic factors, and the effect of mitotane therapy. *N Engl J Med* 1990; 322: 1195–1201.
47. Hague RV, May W, Cullen DR. Hepatic microsomal enzyme induction and adrenal crisis due to o,p'DDD therapy for metastatic adrenocortical carcinoma. *Clin Endocrinol (Oxf)* 1989; 31: 51–57.
48. Schulte HM, Benker G, Reinwein D et al. Infusion of low dose etomidate: correction of hypercortisolemia in patients with Cushing's syndrome and dose-response relationship in normal subjects. *J Clin Endocrinol Metab* 1990; 70: 1426–1430.
49. Preda VA, Sen J, Karavitaki N et al. Etomidate in the Management of Hypercortisolemia in Cushing's Syndrome. *Eur J Endocrinol* 2012; 167: 137–143.
50. Pivonello R, Ferone D, de Herder WW et al. Dopamine receptor expression and function in corticotroph pituitary tumors. *J Clin Endocrinol Metab* 2004; 89: 2452–2462.
51. Boscaro M, Benato M, Mantero F. Effect of bromocriptine in pituitary-dependent Cushing's syndrome. *Clin Endocrinol (Oxf)* 1983; 19: 485–491.
52. Godbout A, Manavela M, Danilowicz K et al. Cabergoline monotherapy in the long-term treatment of Cushing's disease. *Eur J Endocrinol* 2010; 163: 709–716.
53. Vilar L, Naves LA, Azevedo MF et al. Effectiveness of cabergoline in monotherapy and combined with ketoconazole in the management of Cushing's disease. *Pituitary* 2010; 13: 123–129.
54. Day R, Dong W, Panetta R et al. Expression of mRNA for somatostatin receptor (sstr) types 2 and 5 in individual rat pituitary cells. A double labeling in situ hybridization analysis. *Endocrinology* 1995; 136: 5232–5235.
55. Boscaro M, Ludlam WH, Atkinson B et al. Treatment of pituitary-dependent Cushing's disease with the multireceptor ligand somatostatin analog pasireotide (SOM230): a multicenter, phase II trial. *J Clin Endocrinol Metab* 2009; 94: 115–122.
56. Colao A, Peterseim S, Newell-Price J et al. A 12-month phase 3 study of pasireotide in Cushing's disease. *N Engl J Med* 2012; 366: 914–924.
57. Fleseriu M, Biller BM, Findling JW et al. Mifepristone, a Glucocorticoid Receptor Antagonist, Produces Clinical and Metabolic Benefits in Patients with Cushing's Syndrome. *J Clin Endocrinol Metab* 2012; 97: 2039–2049.