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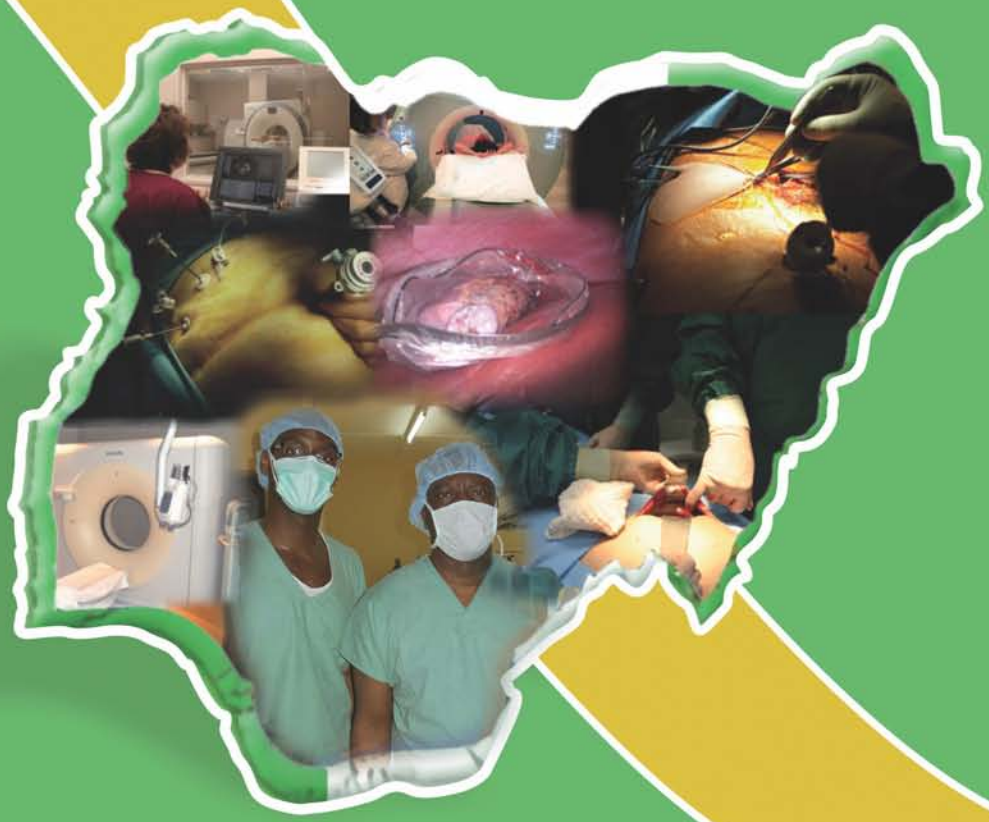

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REVIEW ARTICLE

Urinary free cortisol in the diagnosis of Cushing's syndrome: How useful?

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Abstract

Cushing's Syndrome results from chronic exposure to excessive circulating levels of glucocorticoids. To confirm the clinical suspicion, biochemical tests are needed. These biochemical tests include the measurement of excess total endogenous cortisol secretion assessed by 24-hour urinary free cortisol (UFC), loss of the normal feedback of the hypothalamo-pituitary-adrenal axis assessed by suppressibility after dexamethasone testing, and disturbance of the normal circadian rhythm of cortisol secretion assessed by midnight serum or salivary cortisol. We searched the Medline, Pubmed, journal articles, WHO publications and reputable textbooks relating to Cushing's syndrome using publications from 1995 to 2011. UFC has been the classic screening test used to confirm hypercortisolemia as the first step in diagnostic work-up of Cushing's syndrome. Its long-term use in clinical practice has led to emergence of significant evidence regarding the utility of UFC in the diagnosis of Cushing's syndrome. UFC would have been a simple diagnostic tool to use but for the drawbacks in the sample collection, different laboratory methods of assay, not easily determined normal range. UFC use as a screening test is not strongly favoured because cortisol is not uniformly secreted during the day, and the increased prevalence of mild, preclinical or cyclic Cushing's syndrome. A very high level of UFC negates the need for other test procedures in patients with obvious symptoms and signs of Cushing's syndrome. We therefore suggest that UFC should be used with other screening tests for Cushing's syndrome to increase diagnostic yield.

Key words: Cushing's syndrome, diagnosis, screening, urinary free cortisol

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Introduction

Cushing's Syndrome results from chronic exposure to excessive circulating levels of glucocorticoids.[1] Even now its investigation and management can vex the most experienced endocrinologist. The plethora of investigations often needed for the diagnosis and differential diagnosis has grown over the intervening century, and requires careful interpretation. It is the loss of this circadian rhythm, together with loss of the normal feedback mechanism of the hypothalamo-pituitary-adrenal (HPA) axis, which results in chronic exposure to excessive circulating cortisol levels and that gives rise to the clinical state of endogenous Cushing's syndrome.[2] Hypercortisolemia together with the loss of the normal circadian rhythm of cortisol secretion, and disturbed feedback of the HPA axis, are the cardinal biochemical features of Cushing's syndrome. Tests to confirm the diagnosis are based upon these principles. An excess of total endogenous cortisol secretion is assessed by 24-hour urinary free cortisol (UFC), loss of the normal feedback of the hypothalamo-pituitary-adrenal axis (HPA) is assessed by suppressibility of cortisol secretion after dexamethasone testing (DST), and disturbance of the normal circadian rhythm of cortisol secretion is assessed by midnight serum cortisol (MNC) or late-night salivary cortisol (NSC).[3]

Screening for Cushing's syndrome requires that tests of high sensitivity should be used initially to avoid missing milder cases. Tests of high specificity can then be employed

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to exclude false positives.[2] To make for better sensitivity, threshold of these test have been redefined downwards to a lower value.[4] UFC and the overnight DST have been shown to have the most evidences to both identify and exclude patients with Cushing's syndrome.[5] Is UFC really useful in the diagnosis of Cushing's syndrome? This review focuses on the value of UFC as a screening test for Cushing's syndrome. We searched the Medline, Pubmed, journal articles, WHO publications and reputable textbooks relating to Cushing's syndrome using publications from 1995 to 2011.

Urinary Free Cortisol

Measurement of urinary free cortisol (UFC) is a non-invasive test that is widely used in the screening of Cushing's syndrome. About 8-10% of plasma cortisol is 'free' or unbound and physiologically active. Unbound cortisol is filtered by the kidney, with the majority being reabsorbed in the tubules, and the remainder excreted unchanged. Thus, 24-hour UFC collection produces an integrated measure of serum cortisol, smoothing out the variations in cortisol during the day and night. In a series of 146 patients with Cushing's syndrome, UFC measurement was shown to have a sensitivity of 95% for the diagnosis.[6] Measurement of urinary cortisol is a direct assessment of free circulating (biologically active) cortisol and it is not affected by factors that influence corticosteroid-binding globulin (CBG) levels.[3,7,8] The Consensus Statement for Diagnosis of Cushing's syndrome in 2003 clearly states that UFC values can be extremely variable in Cushing's syndrome.[7] A minimum of three urine collections is thus required when there is high index of suspicion, or when the initial result is normal or to exclude the possibility of cyclic Cushing's syndrome.[7] Cushing's syndrome can be excluded after three normal collections; however values four-fold greater than the upper limit of normal can be considered diagnostic for Cushing's syndrome.[3] Studies have shown that small increases in cortisol production at the circadian nadir may not be detected as an increase in UFC. This is particularly exemplified in patients with mild Cushing's syndrome. They have a small but significant increase in night-time cortisol secretion that does not clearly elevate UFC because most of the cortisol secreted during any 24-hour period is generally between 4:00 and 16:00 hour.[8-10] Vilar L *et al*., in their study found that all the three tests used to confirm hypercortisolemia, except UFC, showed a sensitivity of 100%.[11] It was also discovered that 11.3% of 53 patients had normal UFC values, comparable to the 5-24% in most studies.[2,6,12,13] In the differentiation of Cushing's syndrome from obesity, previous studies have shown that UFC measurement have a diagnostic sensitivity and specificity of 95% and 98%, respectively,[2] but more recent data reported a low sensitivity ranging from 45 to 71% at 100% specificity.[9] These recent results suggest that, though UFC may be useful to confirm Cushing's syndrome, its sensitivity and specificity are not optimal as an initial screening test. Lack of specificity had rendered UFC not to be a useful tool in the assessment of Cushing's syndrome.[14]

Drawbacks of Using Urinary Free Cortisol to Diagnose Cushing's Syndrome

Aside from UFC lacking specificity in the assessment of Cushing's syndrome, there are other factors that render it not very useful in the assessment of Cushing's syndrome.

Laboratory determination of urinary free cortisol

Twenty four-hour urine collection is required for UFC estimation. Simple as it appears, patient education is required using both oral and written instructions. Excessive fluid intake (≥5 L/day) will increase UFC level significantly. Patients should be instructed not to drink excessive amounts of fluid. UFC must be corrected for creatinine concentration to ensure that the urine is adequately collected and complete.[1,7,15] UFC is raised by any physiological or pathological condition that increases cortisol production. Normal result in these conditions is more reliable than an abnormal one. Reproducibility can be low in children and as such at least two collections should be performed.[1] UFC is affected by small impairment in renal function.[7,16] Creatinine clearance of less than 60 ml/min will give a falsely low UFC, and UFC levels fall linearly with more severe renal failure.[1] In renal failure, cortisol metabolism is affected in various ways, including alterations in the HPA axis, a prolonged half-life of serum cortisol, and decreased oxidation of tetrahydrocortisone to tetrahydrocortisol.[16] The recent guidelines consequently recommended the use of 1-mg overnight DST rather than UFC for initial testing for Cushing's syndrome in patients with severe renal failure.[1]

Different analytical methods by laboratories also have the potential to affect UFC.[17,18] Reference ranges from these laboratories are also different. Tandem mass spectroscopy (LC-MS/MS) and separation by high-performance liquid chromatography (HPLC) with subsequent measurement of cortisol by RIA or enzyme-linked immunosorbent assay (ELISA) are the two standard methods for the measurement of UFC at the moment.[1,3,9] Antibody-based immunoassays such as unextracted radioimmunoassay (RIA) and ELISA can be affected by cross reactivity with cortisol metabolites and synthetic glucocorticoids. Structurally based assays such as HPLC and tandem LC-MS/MS do not pose this problem, allowing the separation of various urinary glucocorticoids and metabolites, and are being used with increasing frequency. However, these techniques are more expensive, are not yet widely available, and have not yet been validated extensively.[7] There is substantial variation in normal ranges, depending on the method used. Guidelines have recommended using the upper limit of normal for any particular assay as a positive test.[1]

Urinary free cortisol in pregnancy

Pregnancy is a physiologic state in which there is hypercortisolemia. This is due to the elevated cortisol binding globulin (CBG) which is oestrogen dependent. The plasma cortisol level and adrenocorticotropic hormone level are elevated up to about two to three folds. The UFC is thus elevated in pregnancy state. UFC is normal in the first trimester, but increases through second trimester till term.[19] There is preservation of serum cortisol circadian variation in normal pregnancy, whereas suppression of DST is blunted in pregnancy, and diagnostic thresholds for MNC or NSC in pregnant patients are not known.[1] Guidelines recommend the use of UFC as against the use of DST in the initial evaluation of pregnant women with Cushing's syndrome. The challenge here however, is the determination of what constitutes the normal value.[1] It is however suggested that values greater than three times the upper limit of normal can be taken as suggestive of Cushing's syndrome in pregnancy.[1]

Urinary free cortisol in cushing's syndrome and pseudo-cushing's syndrome

Poorly controlled diabetes mellitus, severe obesity (as opposed to mild obesity where UFC may be reduced), psychiatric disorders (depression, anxiety disorder), polycystic ovarian syndrome, alcoholism and late pregnancy are conditions in which the UFC is mildly elevated. These conditions are known as pseudo-Cushing state.[1,2,7,20,21] It has been postulated that higher brain centres stimulate corticotrophin-releasing hormone (CRH) release in these conditions, with subsequent activation of the entire HPA axis.[1,22] Several tests have been used extensively, however, none has proven fully capable of identifying all cases of Cushing's syndrome. Specifically, differentiating Cushing's syndrome from a pseudo-Cushing state is a major clinical challenge for most endocrinologists.[21] The biochemical picture in Cushing's syndrome and pseudo-Cushing's syndrome are similar, the basal UFC is almost the same.[2] Tirabassi G *et al.* in their study showed that the desmopressin ('DDAVP') test was better in differentiating mild forms of Cushing's disease from pseudo-Cushing's syndrome compared to UFC, DST and MNC. In the same study UFC had the lowest positive predictive value compared to the other tests used.[23]

Urinary free cortisol in mild cushing's syndrome

Guidelines have suggested that the 1-mg DST or NSC, rather than UFC, should be performed in patients suspected of having mild Cushing's syndrome as in the case of adrenal incidentalomas.[1] In a small series by Kidambi S *et al.*, the difficulty in establishing the diagnosis of mild hypercortisolemia in patients without increases in UFC secretion was shown.[24] Seven of the 11 patients with mild Cushing's syndrome did not have an elevation of UFC, and NSC was proven to be more useful in these specific cases. The complementary nature of the three commonly performed initial diagnostic tests was also highlighted in the same study. None of these tests is ideal; if they show one or two normal results mild Cushing's syndrome cannot be excluded in patients with a reasonable index of clinical suspicion.[24]

From the foregoing, it is apparent that NSC, UFC and DST are complementary in the diagnostic evaluation of Cushing's syndrome. These tests may be normal in mild or cyclic Cushing's syndrome leading to missed diagnosis.

Conclusion

UFC has been the archetypal screening test used to confirm hypercortisolemia as the initial diagnostic work-up of Cushing's syndrome. UFC would have been a simple diagnostic tool to use but for the drawbacks in the sample collection, different laboratory methods of assay, not easily determined normal range. A normal UFC does not exclude the diagnosis of Cushing's syndrome. A very high level of UFC negates the need for other test procedures in patients with obvious symptoms and signs of Cushing's syndrome. We therefore suggest that UFC should be used with other screening tests for Cushing's syndrome to increase diagnostic yield. UFC could be combined with dexamethasone suppression test (DST) as an outpatient screening test or midnight serum cortisol (MNC) that assesses the disturbance of the normal circadian rhythm of cortisol secretion as an inpatient screening test.

References

1. Nieman LK, Biller BM, Findling JW, Newell-Price J, Savage MO, Stewart PM, *et al.* The diagnosis of Cushing's syndrome: An Endocrine Society Clinical Practice Guideline. J Clin Endocrinol Metab 2008;93:1526-40.
2. Newell-Price J, Trainer P, Besser M, Grossman A. The diagnosis and differential diagnosis of Cushing's syndrome and pseudo-Cushing's states. Endocr Rev 1998;19:647-72.
3. Newell-Price J, Bertagna X, Grossman AB, Nieman LK. Cushing's syndrome. Lancet 2006;367:1605-17.
4. Pecori Giraldi F, Ambrogio AG, De Martin M, Fatti LM, Scacchi M, Cavagnini F. Specificity of first-line tests for the diagnosis of Cushing's syndrome: assessment in a large series. J Clin Endocrinol Metab 2007;92:4123-9.
5. Elamin MB, Murad MH, Mullan R, Erickson D, Harris K, Nadeem S, *et al.* Accuracy of diagnostic tests for Cushing's syndrome: A systematic review and metaanalyses. J Clin Endocrinol Metab 2008;93:1553-62.
6. Nieman L, Cutler GB Jr. Cushing's syndrome. In: Degroot LJ, Besser M, Burger HG, editors. Endocrinology. 3rd ed. Philadelphia. Pa:WB Saunders; 1995. p. 1741-69.
7. Arnaldi G, Angeli A, Atkinson AB, Bertagna X, Cavagnini F, Chrousos GP, *et al.* Diagnosis and complications of Cushing's syndrome: A consensus statement. J Clin Endocrinol Metab 2003;88:5593-602.
8. Makras P, Toloumis G, Papadogias D, Kaltsas GA, Besser M. The diagnosis and differential diagnosis of endogenous Cushing's syndrome. Hormones (Athens) 2006;5:231-50.
9. Carroll TB, Findling JW. The diagnosis of Cushing's syndrome. Rev Endocr Metab Disord 2010;11:147-53.
10. Findling JW, Raff H. Cushing's Syndrome: Important issues in diagnosis and management. J Clin Endocrinol Metab 2006;91:3746-53.
11. Vilar L, Freitas MC, Naves LA, Canadas V, Albuquerque JL, Botelho CA, *et al.* The role of non-invasive dynamic tests in the diagnosis of Cushing's syndrome. J Endocrinol Invest 2008;31:1008-13.

12. Lin CL, Wu TJ, Machacek DA, Jiang NS, Kao PC. Urinary free cortisol and cortisone determined by high performance liquid chromatography in the diagnosis of Cushing's syndrome. J Clin Endocrinol Metab 1997;82:151-5.
13. Vilar L, Naves LA, Freitas Mda C, Moura E, Canadas V, Leal E, *et al.* [Endogenous Cushing's syndrome: Clinical and laboratorial features in 73 cases]. Arq Bras Endocrinol Metabol 2007;51:566-74.
14. Pearson Murphy BE. Lack of specificity of urinary free cortisol determinations: Why does it continue? J Clin Endocrinol Metab 1999;84:2258-9.
15. Shi L, Wudy SA, Maser-Gluth C, Hartmann MF, Remer T. Urine volume dependency of specific dehydroepiandrosterone (DHEA) and cortisol metabolites in healthy children. Steroids 2011;76:140-4.
16. Issa BG, Page MD, Read G, John R, Douglas-Jones A, Scanlon MF. Undetectable urinary free cortisol concentrations in a case of Cushing's disease. Eur J Endocrinol 1999;140:148-51.
17. Persichilli S, Gervasoni J, Iavarone F, Zuppi C. A simple liquid chromatography-tandem mass spectrometry method for urinary free cortisol analysis: Suitable for routine purpose. Clin Chem Lab Med 2010;48:1433-7.
18. Oledzka I, Plenis A, Konieczna L, Kowalski P, Baczek T. Micellar electrokinetic chromatography for the determination of cortisol in urine samples in view of biomedical studies. Electrophoresis 2010;31:2356-64.
19. Vilar L, Freitas Mda C, Lima LH, Lyra R, Kater CE. Cushing's syndrome in pregnancy: An overview. Arq Bras Endocrinol Metabol 2007;51:1293-302.
20. Vilar L, Freitas Mda C, Faria M, Montenegro R, Casulari LA, Naves L, *et al.* Pitfalls in the diagnosis of Cushing's syndrome. Arq Bras Endocrinol Metabol 2007;51:1207-16.
21. Boscaro M, Arnaldi G. Approach to the patient with possible Cushing's syndrome. J Clin Endocrinol Metab 2009;94:3121-31.
22. Krikorian A, Khan M. Is metabolic syndrome a mild form of Cushing's syndrome? Rev Endocr Metab Disord 2010;11:141-5.
23. Tirabassi G, Faloia E, Papa R, Furlani G, Boscaro M, Arnaldi G. Use of the desmopressin test in the differential diagnosis of pseudo-Cushing state from Cushing's disease. J Clin Endocrinol Metab 2010;95:1115-22.
24. Kidambi S, Raff H, Findling JW. Limitations of nocturnal salivary cortisol and urine free cortisol in the diagnosis of mild Cushing's syndrome. Eur J Endocrinol 2007;157:725-31.

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