



SALIVA AND BLOOD SPOT CORTISOL TESTING FOR ADRENAL FUNCTION

The Problem

People in today's society tend to lead frenetic, unbalanced lifestyles. Commonly, individuals experience continuous stress, not only from emotional stressors (e.g. marital, financial, and occupational) but also from physical stressors (e.g. sleep deprivation, caffeine consumption, pain, extreme exercise) without adequate recovery. Chronic exposure to these stressors often causes elevations in adrenal hormone levels, leading to disorders ranging from anxiety to infertility. While many individuals are able to cope, the pituitary gland may, over time, decrease the ACTH signal to the adrenal glands that creates an impaired adrenal response to stressors, which reduces adrenal hormone output. The resulting hypocortisolism may present with a constellation of symptoms from chronic fatigue to allergies. While everyone is potentially at risk, the problem is more prevalent among people with high-stress professions (e.g. medical professionals, police officers, executives and teachers).

Adrenal Function and Cortisol

The zona fasciculata of the adrenal cortex secretes approximately 15-20 mg of cortisol per day. Under the direction of the hypothalamus and pituitary and controlled by a negative feedback loop, the zona fasciculata is stimulated by adrenocorticotrophic hormone (ACTH) to produce cortisol in response to stressors. This feedback loop is commonly referred to as the hypothalamic-pituitary-adrenal (HPA) axis. Cortisol has a wide range of effects on mind and body and interacts with the reproductive, thyroid and immunological systems. As part of the response to stress, it prepares the body for "fight or flight" and in doing so it can suppress the production of other hormones. This temporarily shuts down processes that would otherwise divert the body's resources away from its more immediate requirement to respond to the stressor, including processes involved with reproduction and some immune functions. Because of these effects, when cortisol levels remain chronically high, this suppression of other processes is maintained for longer than normal and this can result in susceptibility to infection, hypothyroidism, bone loss, and low libido. On the other hand, lower than normal cortisol levels are associated with decreasing attention span, fatigue, and blood sugar imbalances. Since both high and low cortisol levels are associated with multiple symptoms, cortisol

testing often provides the answers to complicated health situations, which have led patients to visit multiple physicians without success.

Successful diagnosis and treatment of the underlying problem improves patient symptoms, and cortisol testing provides an objective measurement of response to treatment. There are several good books and websites that discuss the relationship between adrenal function, cortisol levels and overall health, and some of these are listed under Useful Resources at the end of this sheet.

Blood Spot and Saliva Cortisol Testing

Salivary cortisol testing is an established method for the diagnosis of Cushing's Disease (hypercortisolism) and preferred over serum or urine testing because of its reliability, non-invasiveness and convenience of sampling¹. In research on HPA axis function, salivary cortisol is also preferred to serum measurements as a reliable indicator of adrenal status because it represents the free, bioavailable hormone levels, excluding the cortisol-binding globulin (CBG)-bound hormone that circulates in the blood but does not pass into saliva²⁻⁶. The convenience of saliva collection and the avoidance of an anticipatory rise in cortisol levels caused by the stress of venipuncture has also been an advantage in research on adrenal function and depression in infants and children^{7,8}. Saliva collection is ideal for multiple sampling over the course of a day and it is therefore the medium most commonly used in clinical studies of adrenal function, for example the study of job-related stress in professionals such as teachers^{9,10}.

Blood spot testing is also minimally-invasive, involving just a momentary finger-stick and collection of a few drops of blood on a filter paper. Blood spot cortisol levels have been found to correlate well with serum levels¹¹ and the blood spot test for morning cortisol gives an excellent snapshot of adrenal function without the stress and inconvenience of venipuncture serum collection.

Cortisol Awakening Response (CAR)

In a process governed by the hippocampus and the light-sensitive suprachiasmatic nucleus of the hypothalamus, cortisol levels rise significantly between the moment of waking

and 30 or so minutes after rising, returning to the initial waking level at 60 minutes after waking. The magnitude of this cortisol awakening response (CAR) gives us some idea of the HPA axis's ability to activate and respond to stress and gives us even more clues to pin down the reason for a hyperactive or hypoactive HPA axis. This transient rise in cortisol levels is unique to the stimulus of morning waking. This phenomenon does not occur when woken in the middle of the night or after an afternoon nap. Further, the findings tend to be consistent sample to sample for a given person regardless of the first waking cortisol. So, if the CAR is tested one day and then on a day a week later, the extent of the rise should be the same. This could be a very helpful stake in the sand given the fact that morning cortisol levels have a comparatively higher intra-individual variation from day to day. A 50% increase in cortisol from the time of waking to 30 minutes later is considered a normal CAR^{12,13}.

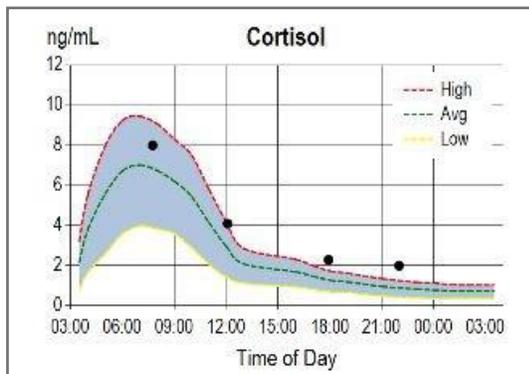
Testing Options

ZRT offers a single morning cortisol test in either saliva or blood spot, and this is included in all our multi-hormone profiles because of the interconnection of adrenal function with other hormone systems.

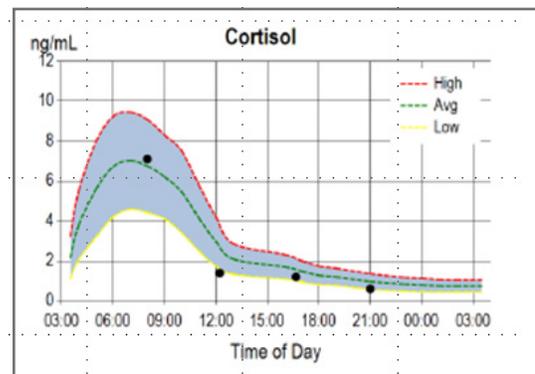
Because of the additional information to be gained by observing a patient's diurnal pattern, saliva tests are offered for twice a day sampling (morning and bedtime), or four times during a day (within one hour of waking, mid-day, evening and bedtime). The Adrenal Stress Profile kit includes 6 collection tubes and the test requisition allows the provider to choose either a standard diurnal collection or the diurnal cortisol plus CAR.

Diurnal Patterns of Cortisol Production

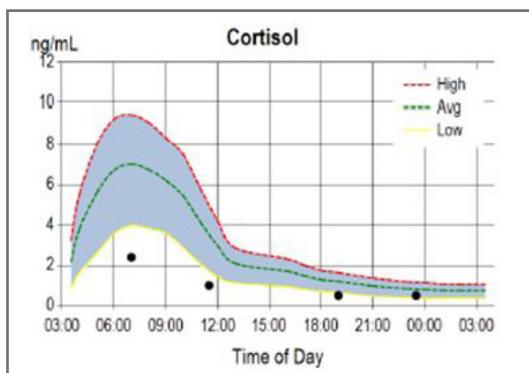
Cortisol production is normally at its highest 30 minutes after waking and declines steadily during the day, reaching its lowest point at bedtime. Examples of some patterns that can be seen in various types of cortisol dysfunction are shown below, against a shaded area showing the normal range at each point during the day. ZRT test reports show cortisol test results plotted at the actual time of day when samples are collected, for both saliva and blood spot. The examples below are in saliva.



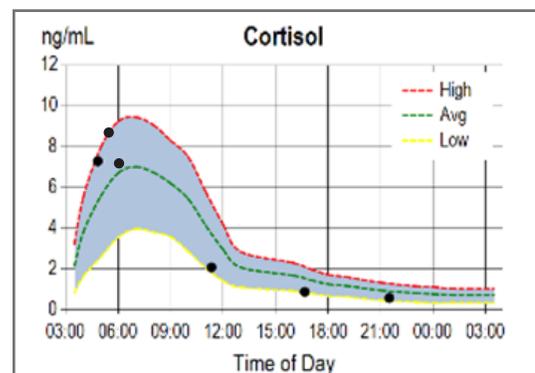
Chronic adrenal stress pattern – overall higher than normal cortisol production throughout the day



Stress/fatigued pattern – morning cortisol in the high normal range or elevated, but levels drop off rapidly, indicating adrenal dysfunction



Hypocortisolism/burnout pattern – morning cortisol surge is suppressed and overall diurnal pattern is flattened



Normal circadian rhythm with cortisol awakening response

References

1. Trilck M, Flitsch J, Lüdecke DK, Jung R, Petersenn S. Salivary cortisol measurement--a reliable method for the diagnosis of Cushing's syndrome. *Exp Clin Endocrinol Diabetes*. 2005;113:225-30. Erratum in: *Exp Clin Endocrinol Diabetes*. 2005 Jul;113(7):409.
2. Aardal E, Holm AC. Cortisol in saliva – reference ranges and relation to cortisol in serum. *Eur J Clin Chem Clin Biochem* 1995;33:927-32.
3. Aardal-Eriksson E, Karlberg BE, Holm AC. Salivary cortisol – an alternative to serum cortisol determinations in dynamic function tests. *Clin Chem Lab Med* 1998;36:215-22.
4. Gozansky WS, Lynn JS, Laudenslager ML, Kohrt WM. Salivary cortisol determined by enzyme immunoassay is preferable to serum total cortisol for assessment of dynamic hypothalamic-pituitary-adrenal axis activity. *Clin Endocrinol (Oxf)*. 2005;63:336-41.
5. Lo MS, Ng ML, Azmy BS, Khalid BA. Clinical applications of salivary cortisol measurements. *Singapore Med J* 1992;33:170-3.
6. Westermann J, Demir A, Herbst V. Determination of cortisol in saliva and serum by a luminescence-enhanced enzyme immunoassay. *Clin Lab* 2004;50:11-24.
7. Goodyer IM, Herbert J, Altham PM, Pearson J, Secher SM, Shiers HM. Adrenal secretion during major depression in 8- to 16-year olds, I. Altered diurnal rhythms in salivary cortisol and dehydroepiandrosterone (DHEA) at presentation. *Psychol Med* 1996;26:245-256.
8. Kiess W, Meidert A, Dressendörfer RA, Schriever K, Kessler U, König A, Schwarz HP, Strasburger CJ. Salivary cortisol levels throughout childhood and adolescence: relation with age, pubertal stage, and weight. *Pediatr Res* 1995;37:502-6.
9. Pruessner JC, Hellhammer DH, Kirschbaum C. Burnout, perceived stress, and cortisol responses to awakening. *Psychosom Med* 1999;61:197-204.
10. Steptoe A, Copley M, Griffith J, Kirschbaum C. Job strain and anger expression predict early morning elevation in salivary cortisol. *Psychosom Med* 2000;62:286-92.
11. Wong V, Yan T, Donald A, McLean M. Saliva and bloodspot cortisol: novel sampling methods to assess hydrocortisone replacement therapy in hypoadrenal patients. *Clin Endocrinol (Oxf)* 2004;61:131-7.

12. Stalder T, Kirschbaum C, Kudielka BM, Adam EK, Pruessner JC, Wüst S, Dockray S, Smyth N, Evans P, Hellhammer DH, Miller R, Wetherell MA, Lupien SJ, Clow A. Assessment of the cortisol awakening response: Expert consensus guidelines. *Psychoneuroendocrinology*. 2016;63:414-32.
13. Fries E, Dettenborn L, Kirschbaum C. The cortisol awakening response (CAR): facts and future directions. *Int J Psychophysiol*. 2009;72:67-73.

Useful Resources:

Thomas G. Guilliams. *The Role of Stress and the HPA Axis in Chronic Disease Management*. Point Institute; 2015.

James L. Wilson. *Adrenal Fatigue: The 21st Century Stress Syndrome*. Smart Publications; 2001.

www.adrenalfatigue.org

Shawn M. Talbott. *The Cortisol Connection: Why Stress Makes You Fat And Ruins Your Health – And What You Can Do About It*. Hunter House Inc.; 2002.

Cherie Carter-Scott. *If Life Is A Game, These Are The Rules*. Broadway Books; 1998.

Richard Shames, Karilee Shames. *Feeling Fat, Fuzzy or Frazzled? A 3-Step Program to: Beat Hormone Havoc, Restore Thyroid, Adrenal, and Reproductive Balance, and Feel Better Fast!* Hudson Street Press; 2005.

www.feelingfff.com