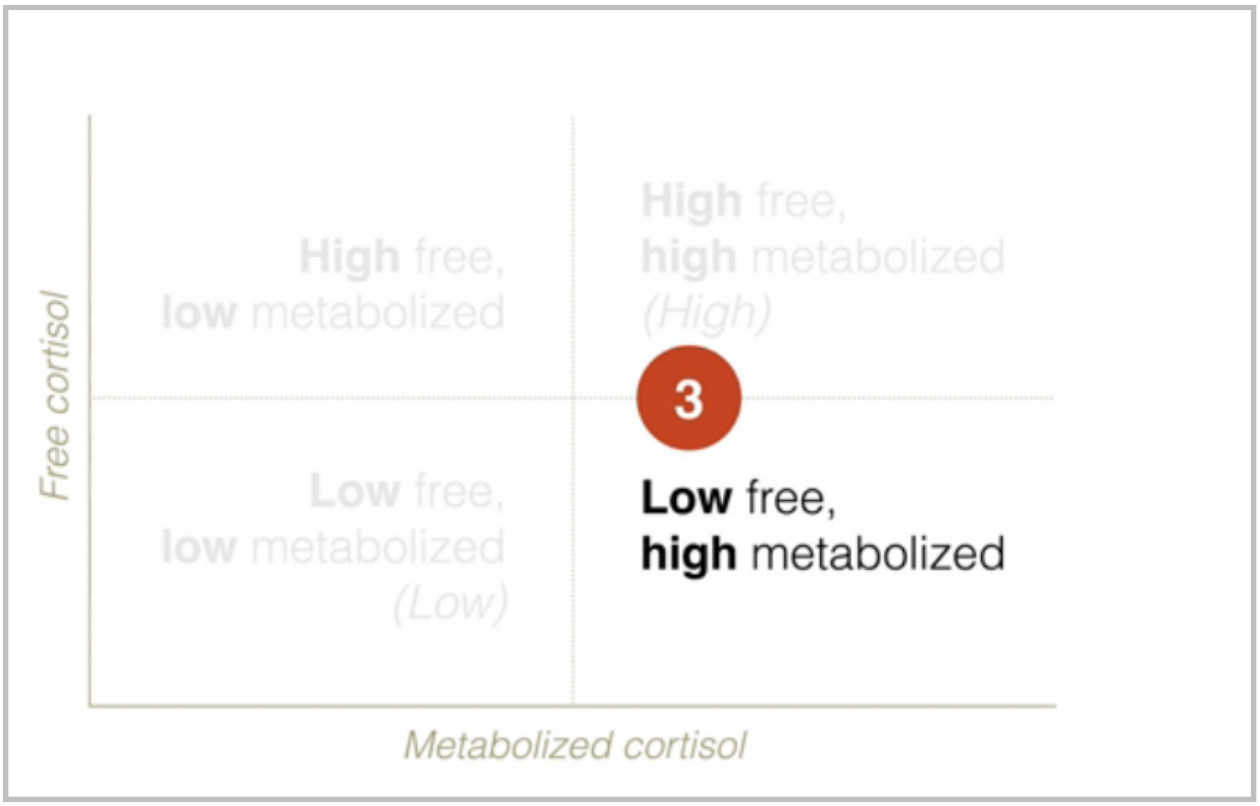
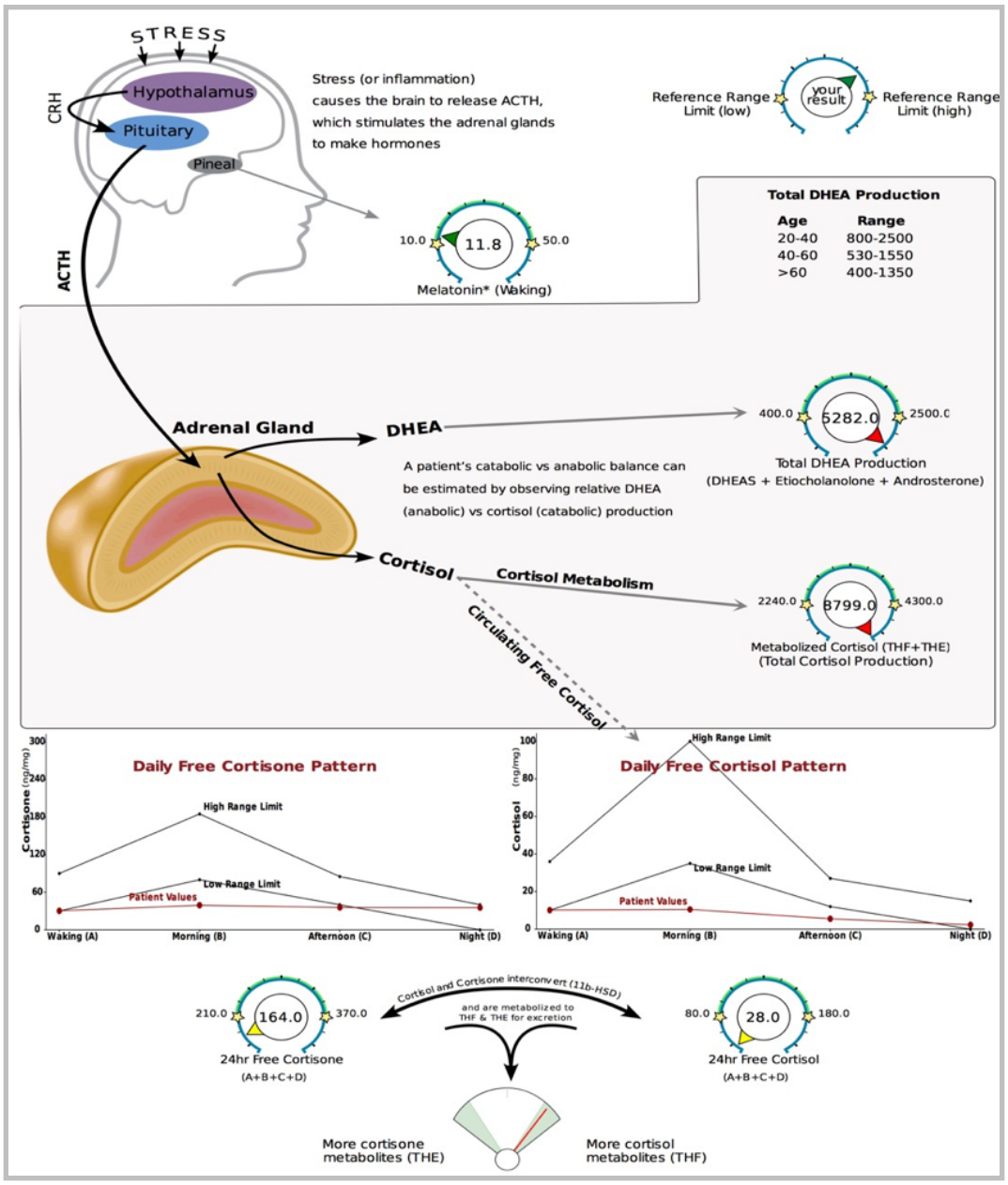
HPA-D: DUTCH Test I - Part 5

Okay, moving on to pattern number three.

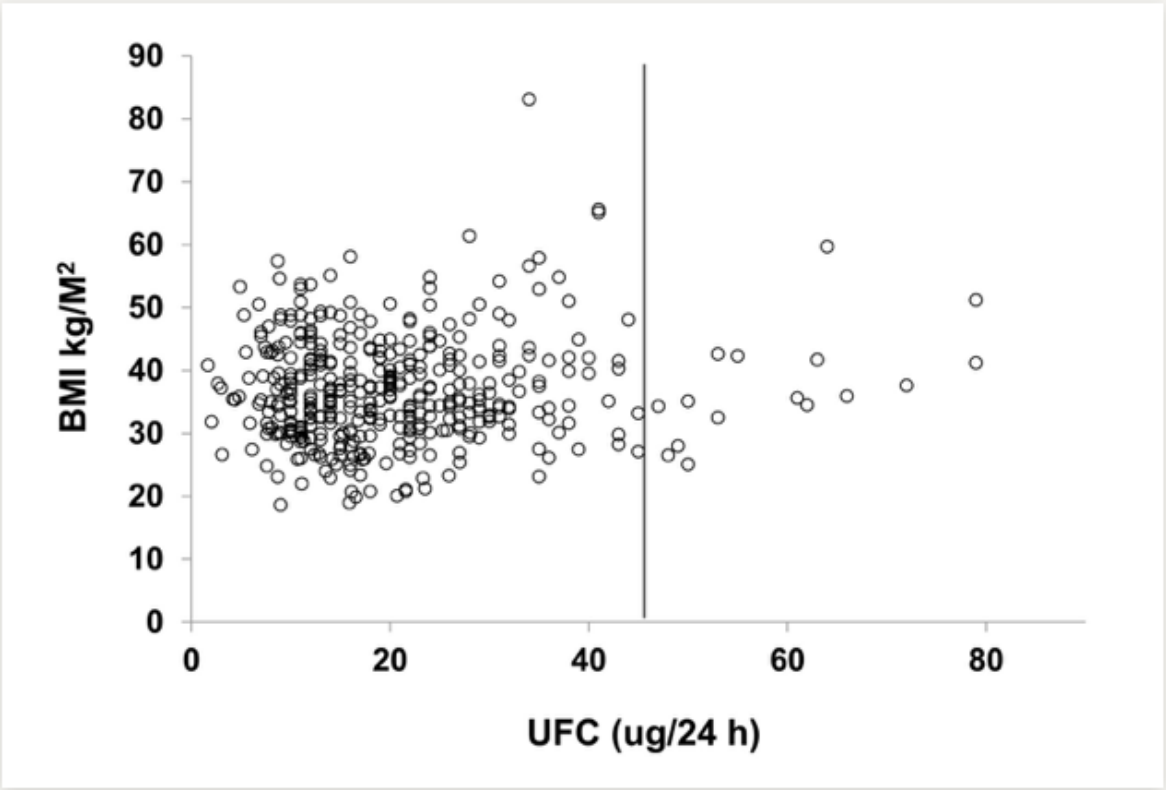


This is when we start with the discordant pattern, so low free cortisol but high total or metabolized cortisol. There are several potential causes of this, but the main ones are obesity, insulin resistance and other metabolic dysfunction, hyperthyroidism, chronic stress, glucocorticoid use, and chronic fatigue syndrome.

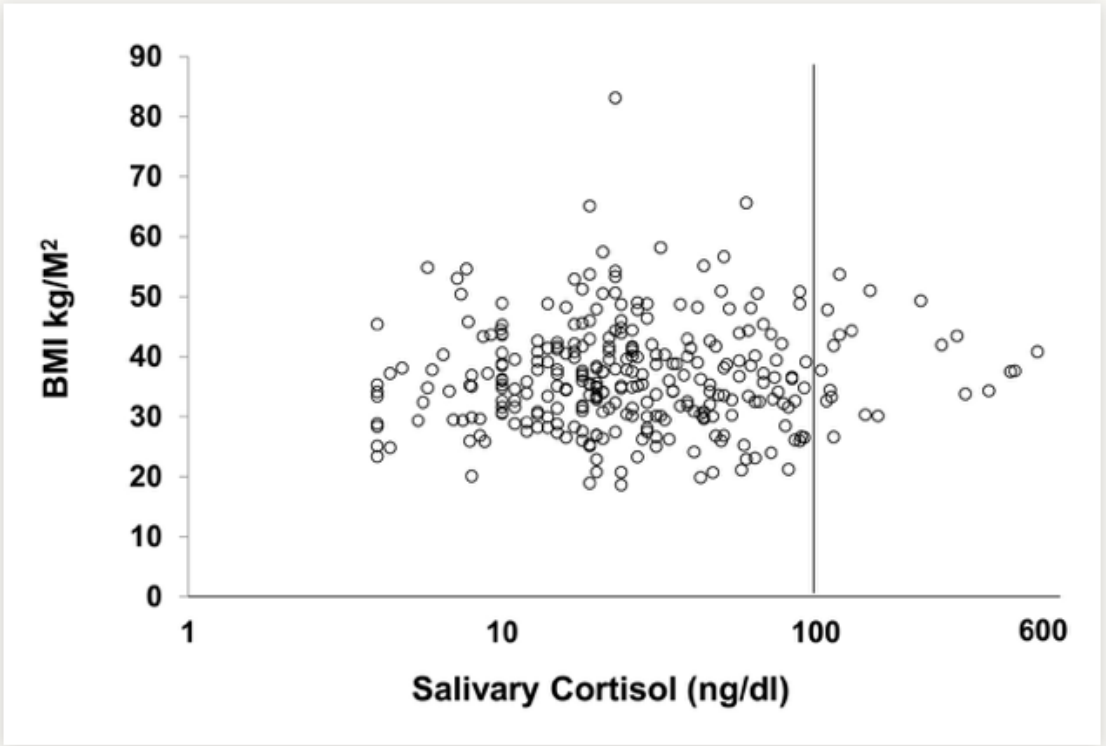


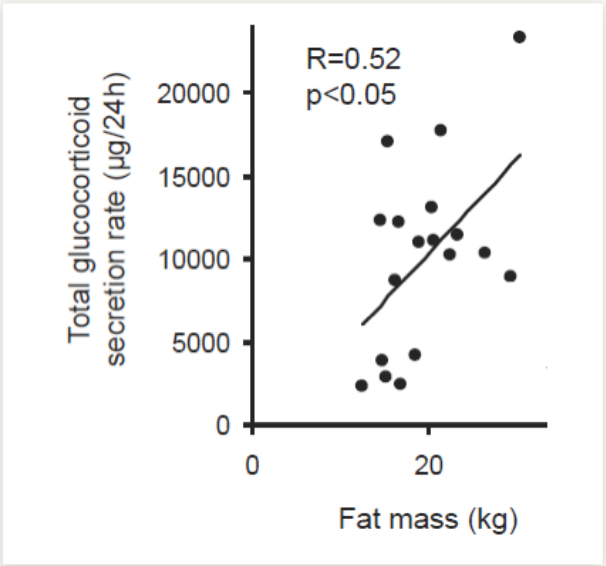
So here we see very low free cortisol with a flattened cortisol rhythm. It's not low enough to trigger a concern about Addison's, but if only free cortisol were measured, this patient would certainly be diagnosed with adrenal fatigue. If you look at this patient's cortisol metabolites, they are almost at 9,000, over two times the upper end of the range, and DHEA is at 5,282, which is over two times the upper end of the range as well. So this patient was 420 pounds with significant insulin and leptin resistance, and this is obesity, not adrenal fatigue.

Urine free cortisol



Saliva free cortisol





Abraham et al. Obesity (Silver Spring). 2013 Jan;21(1):E105-17

This has been studied extensively in the literature. In obesity, we'll typically see normal or low free cortisol, and then we'll see high cortisol metabolites. So the figures on this slide are from a study in the journal ***Obesity***, which examined the relationship between urinary free cortisol, saliva free cortisol, and obesity. As you can see on the upper left there, there is really no correlation between BMI and free cortisol in the saliva or urine, but when you look at the metabolites, there is a linear increase in cortisol metabolites as BMI increases.


ORIGINAL ARTICLE

Reduced Glucocorticoid Production Rate, Decreased 5α-Reductase Activity, and Adipose Tissue Insulin Sensitization After Weight Loss

Jeremy W. Tomlinson,[1] Joanne Finney,[2] Beverly A. Hughes,[1] Susan V. Hughes,[1] and Paul M. Stewart[1]

OBJECTIVE—The epidemics of obesity, insulin resistance, and type 2 diabetes have heightened the need to understand mechanisms that contribute to their pathogenesis. Increased endogenous glucocorticoid production has been implicated based on parallels with Cushing's syndrome. We have assessed the impact of weight loss on glucocorticoid secretion and metabolism (notably 11β-hydroxysteroid dehydrogenase type 1 and 5α-reductase [5αR] activity) and insulin sensitivity.

RESEARCH DESIGN AND METHODS—Twenty obese volunteers were investigated before and after weight loss. Patients underwent hyperinsulinemic-euglycemic clamps with simultaneous adipose microdialysis and oral cortisone acetate administration. Changes in glucocorticoid secretion and metabolism were assessed using 24-h urine collections.

RESULTS—Before weight loss, fat mass correlated with glucocorticoid secretion rate (total fat, $r = 0.46$, $P < 0.05$; trunk fat, $r = 0.52$, $P < 0.05$); however, glucocorticoid secretion rate was inversely related to insulin sensitivity ($r = -0.51$, $P < 0.05$). Hyperinsulinemia failed to suppress adipose tissue interstitial fluid glycerol release (180 ± 50 μmol [basal] vs. 153 ± 10 μmol [steady state], NS). After oral cortisone (25 mg), cortisol concentrations within adipose interstitial fluid increased (4.3 ± 1.1 vs. 14.2 ± 2.6 nmol/l, $P < 0.01$), but glycerol concentrations did not change. After weight loss, insulin sensitivity increased. Consistent with insulin sensitization, adipose tissue interstitial fluid glycerol concentrations fell under hyperinsulinemic conditions (186 ± 16 vs. 117 ± 9 μmol, $P < 0.05$). Glucocorticoid secretion decreased (11,751 ± 1,520 vs. 7,464 ± 937 μg/24 h, $P < 0.05$) as did 5αR activity (5α-tetrahydrocortisol-to-tetrahydrocortisol ratio 1.41 ± 0.16 vs. 1.12 ± 0.17, $P < 0.005$).

CONCLUSIONS—Obesity is associated with insulin resistance within adipose tissue and increased cortisol secretion rates; both are reversed with weight loss. Reduced 5αR activity after weight loss may decrease hypothalamo-pituitary-adrenal axis activation and reduce glucocorticoid metabolite production. ***Diabetes* 57: 1536–1543, 2008**

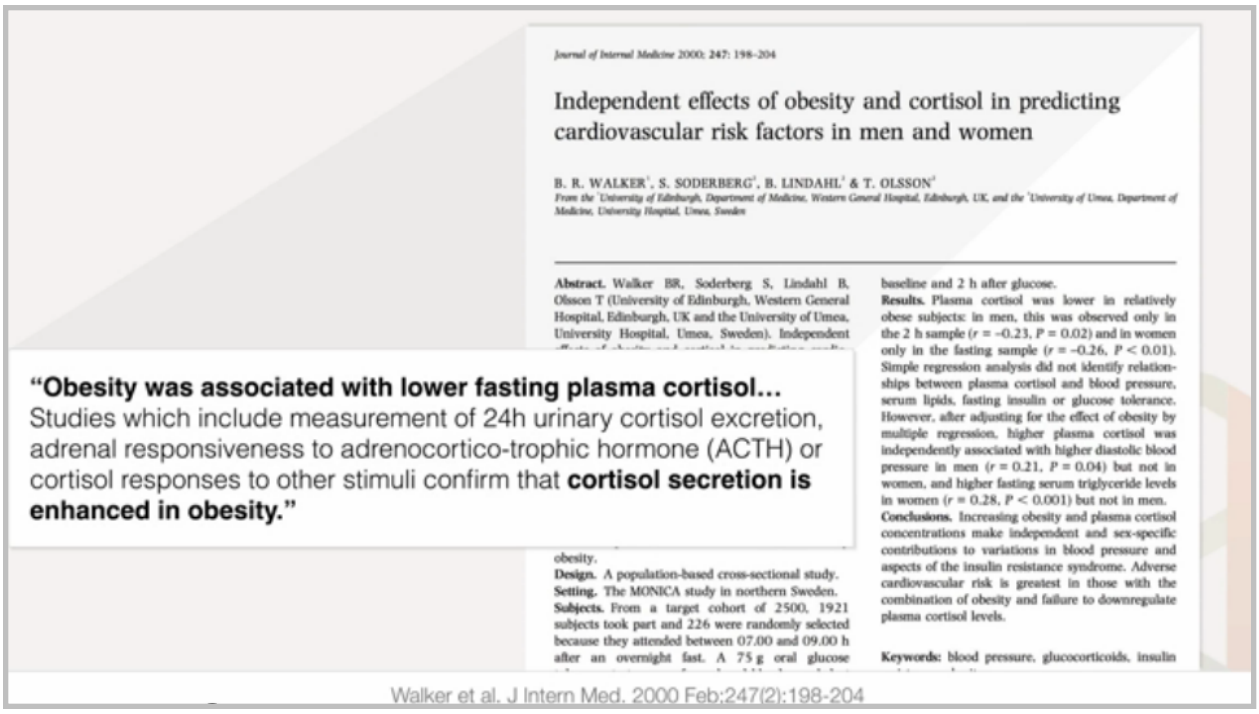
The global epidemic of obesity continues to progress at an alarming rate in both adults and children (1). The health consequences of obesity are well described along with significant increases in both mortality and morbidity (2). The risks of type 2 diabetes and insulin resistance associated with

(11β-HSD1) is highly expressed in liver and adipose tissue (8), and its predominant activity is to convert the inactive glucocorticoid cortisone to active cortisol and thereby locally amplify the action of glucocorticoids. In contrast, the A-ring reductases (5α- and 5β-reductase [5αR and 5βR]) inactivate cortisol (in conjunction with 3α-hydroxysteroid dehydrogenase) to its tetrahydro-metabolites (5α-tetrahydrocortisol [5αTHF] and tetrahydrocortisone [THE]). The activities of both these enzyme systems can limit glucocorticoid availability and also impact on HPA

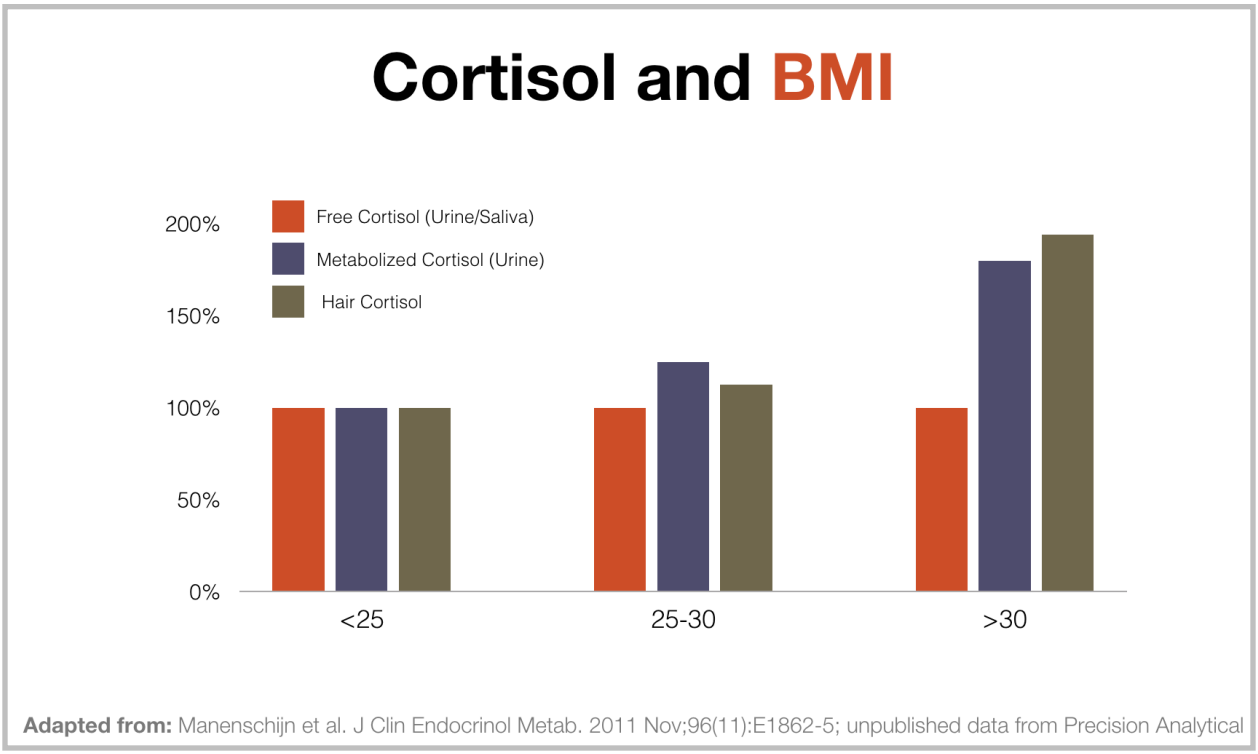
"The sum of total cortisol metabolites provides a reflection of cortisol secretion rate… **After weight loss, total glucocorticoid secretion decreased."**
(Metabolites by 30%, but free cortisol by only 10%)

Tomlinson et al. Diabetes. 2008 Jun;57(6):1536-43

The study on this slide found that cortisol metabolites are the best indicator of total cortisol production. The authors concluded that "the sum of total cortisol metabolites provides a reflection of cortisol secretion rate. After weight loss, total glucocorticoid secretion decreased," and the metabolites decreased by 30 percent, but free cortisol only went down by 10 percent.



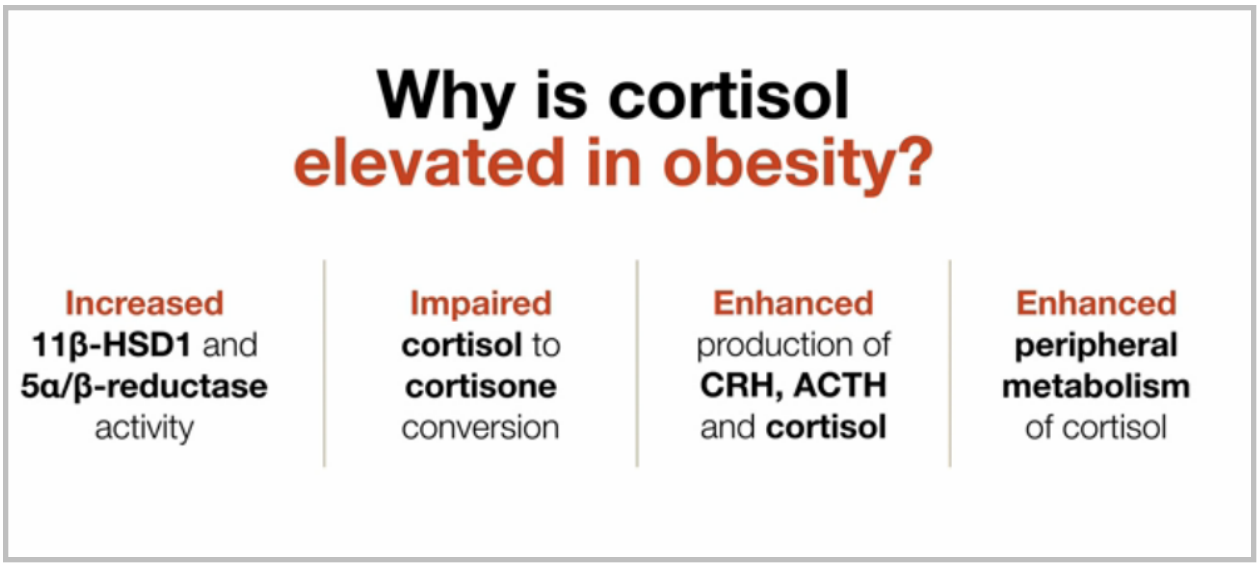
The study on this slide showed that obesity is associated with low fasting plasma cortisol, but it confirms other research showing that obese subjects have higher levels of cortisol metabolites.



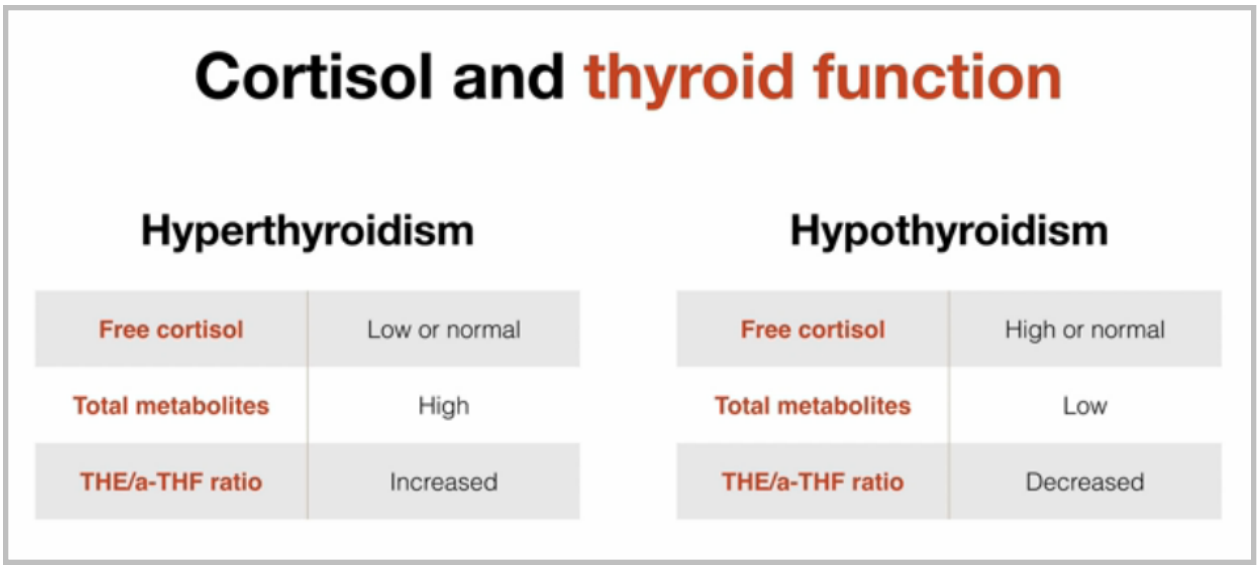
Adapted from: Manenschijn et al. J Clin Endocrinol Metab. 2011 Nov;96(11):E1862-5; unpublished data from Precision Analytical

As you can see on this chart, there is no correlation between free cortisol, whether in the urine or saliva, and body mass index, but there is a positive correlation between cortisol in the hair and metabolized cortisol in the urine and body mass index, and these are both better indicators of overall cortisol production.

The data on this slide come from the ***JCEM*** hair cortisol paper and from internal data from Precision Analytical on saliva and urine tests, and we'll provide a link to the study in the resources section.



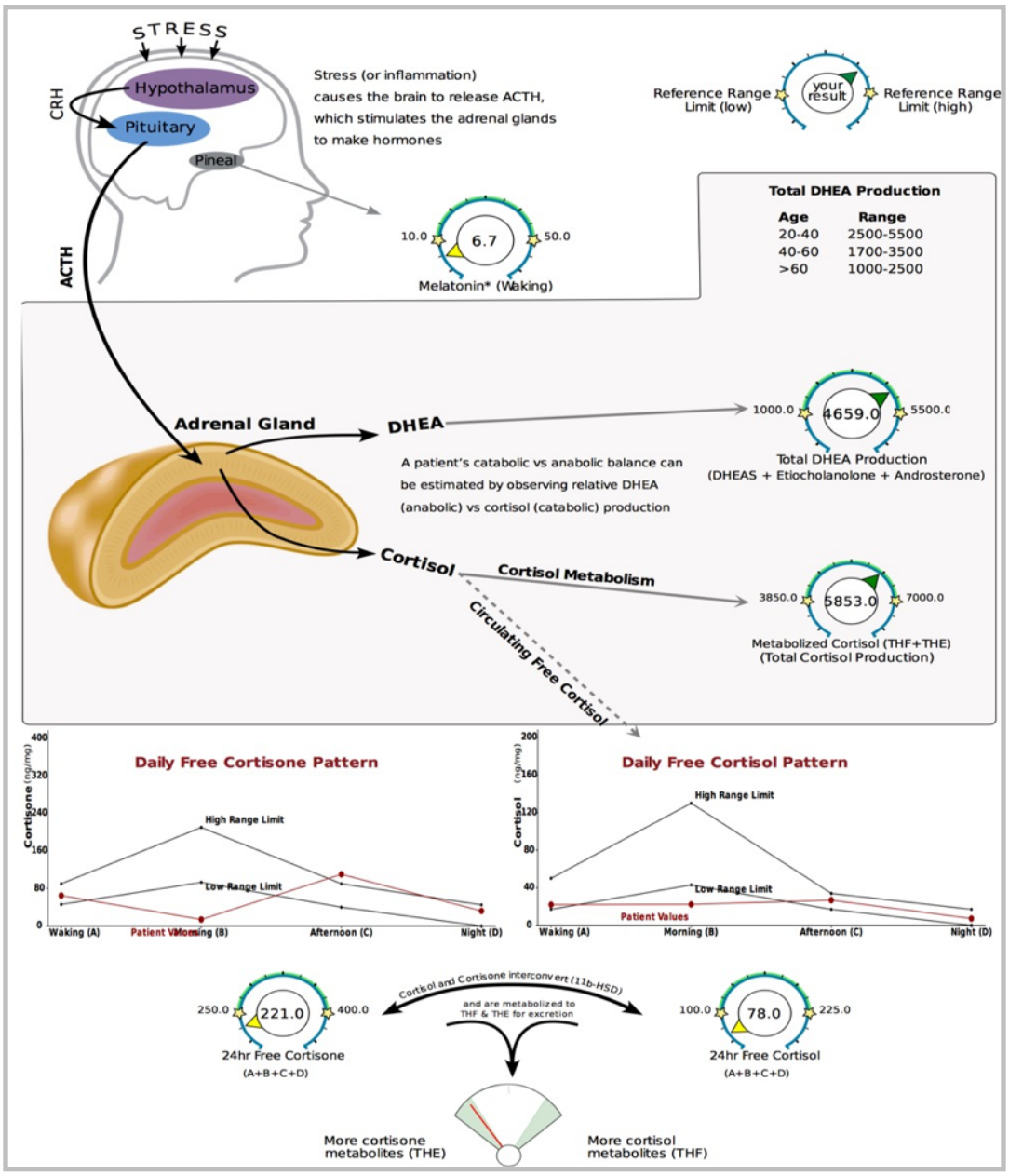
So why are total metabolized cortisol and DHEA elevated in obesity? It's not entirely clear yet in the research, but several possibilities have been observed. It's been shown that fat cells make cortisol via 11β-HSD1 and localized 5α- or 5β-reductase activity, which would mean that more fat equals more cortisol in the tissues. Cortisol-to-cortisone conversion is impaired in obesity, and cortisone shows up in the urine metabolites. It may be a neuroendocrine abnormality that enhances central production of CRH, ACTH, and cortisol. Peripheral metabolism of cortisol may be enhanced, and DHEA is 10 times higher in fat cells than it is in the circulation.



Studies have also shown that cortisol metabolites like tetrahydrocortisone, THE, and allo-tetrahydrocortisol, which is ATHF, are significantly increased in hyperthyroidism. It has also been shown that the ratio of tetrahydrocortisone to allo-tetrahydrocortisol is elevated in hyperthyroidism, and this is thought to be by increases in 11β-HSD and 5α-reductase activity, which impacts peripheral cortisol metabolism.

So I've put a summary of expected findings for cortisol in both hyperthyroidism and hypothyroidism here on the slide. So in hyperthyroidism, we'd expect to see low or normal free cortisol and high cortisol metabolites, and we'd expect to see an increased ratio of tetrahydrocortisone to allo-tetrahydrocortisol. In hypothyroidism, we'd expect to see high or normal free cortisol, low cortisol metabolites, and a decreased ratio of tetrahydrocortisone to allo-tetrahydrocortisol.

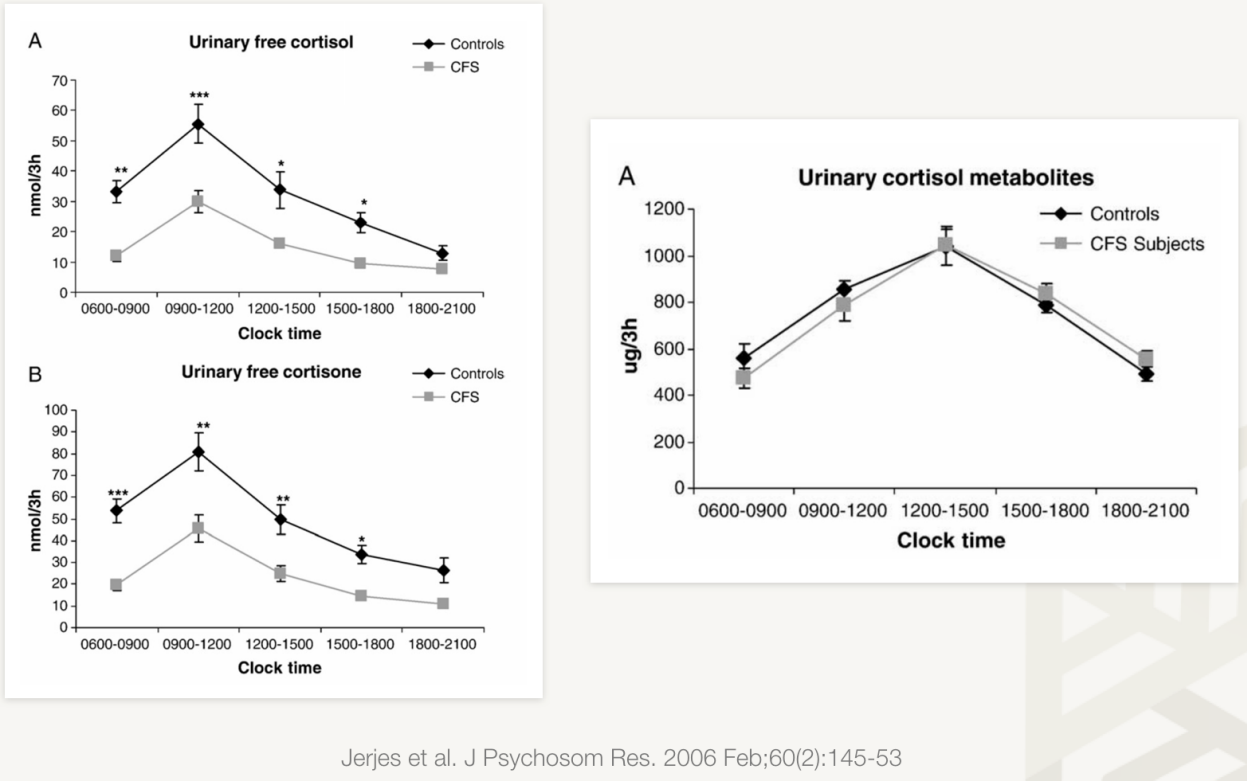
Some researchers have even suggested using urinary cortisol metabolite measurements as a means of diagnosing subclinical thyroid dysfunction, both hypothyroidism and hyperthyroidism. So this is yet another reason why I prefer the DUTCH test because it can pick up on things like this that would be completely missed with a saliva test.



Here is a patient with low free cortisol, low free cortisone, and a disrupted diurnal cortisol rhythm, but metabolites are high normal. This patient is not obese or insulin resistant, is not taking exogenous glucocorticoids, and is not hyperthyroid, but this patient did have an active stress response. She had heavy metal toxicity, SIBO, and depression as primary complaints, so in this case, the pattern is due to chronic stress, not due to obesity, insulin resistance, or hyperthyroidism.

It is well established that exogenous steroid use suppresses the HPA axis and reduces the endogenous production of cortisol, as we've discussed already. In these situations, sometimes you see low free cortisol and low cortisol metabolites, but other times you'll see low free cortisol, but

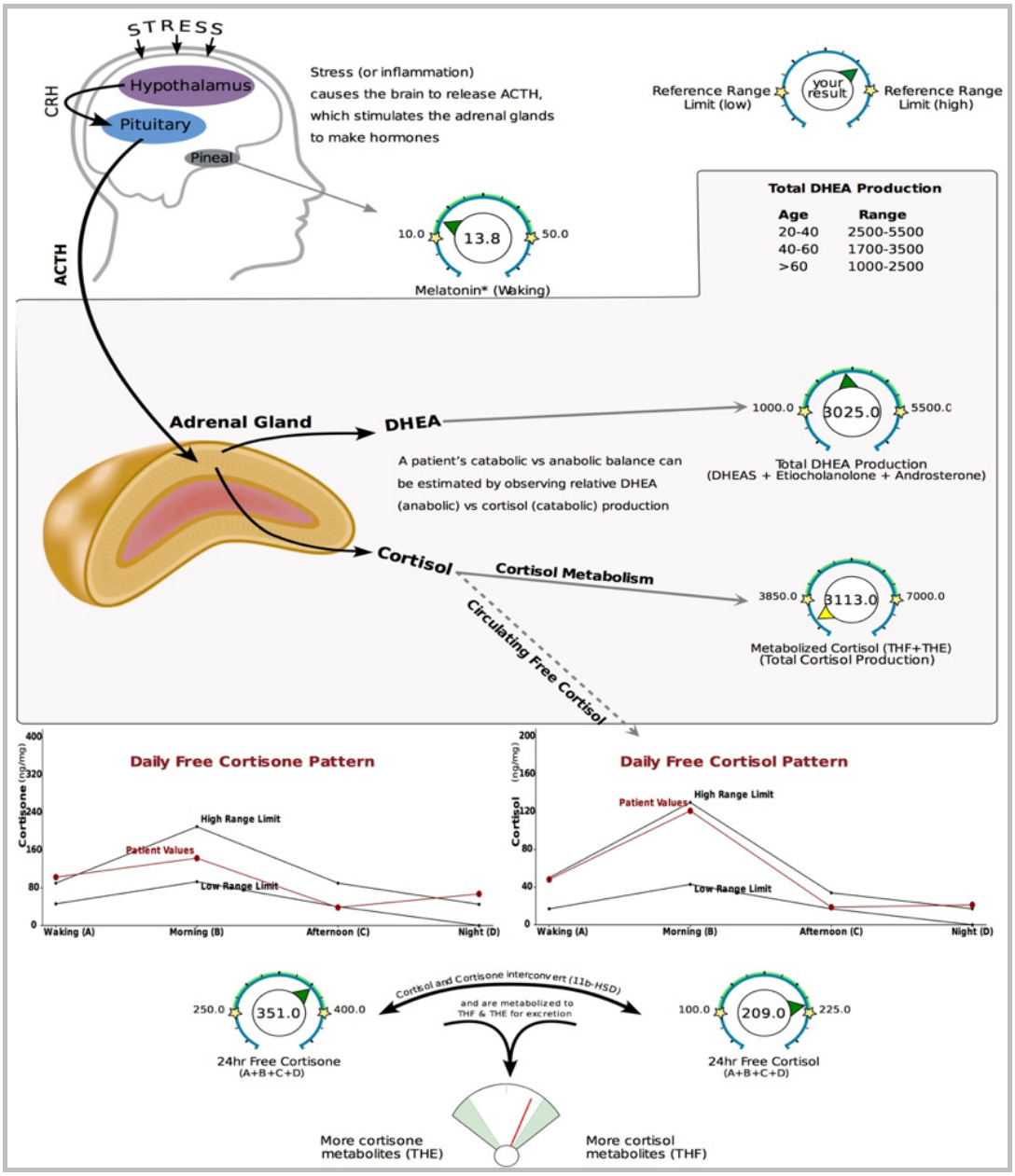
you'll see urinary cortisol metabolites that are normal or even high because they are excreting excess glucocorticoids from the medication.



Jerjes et al. J Psychosom Res. 2006 Feb;60(2):145-53

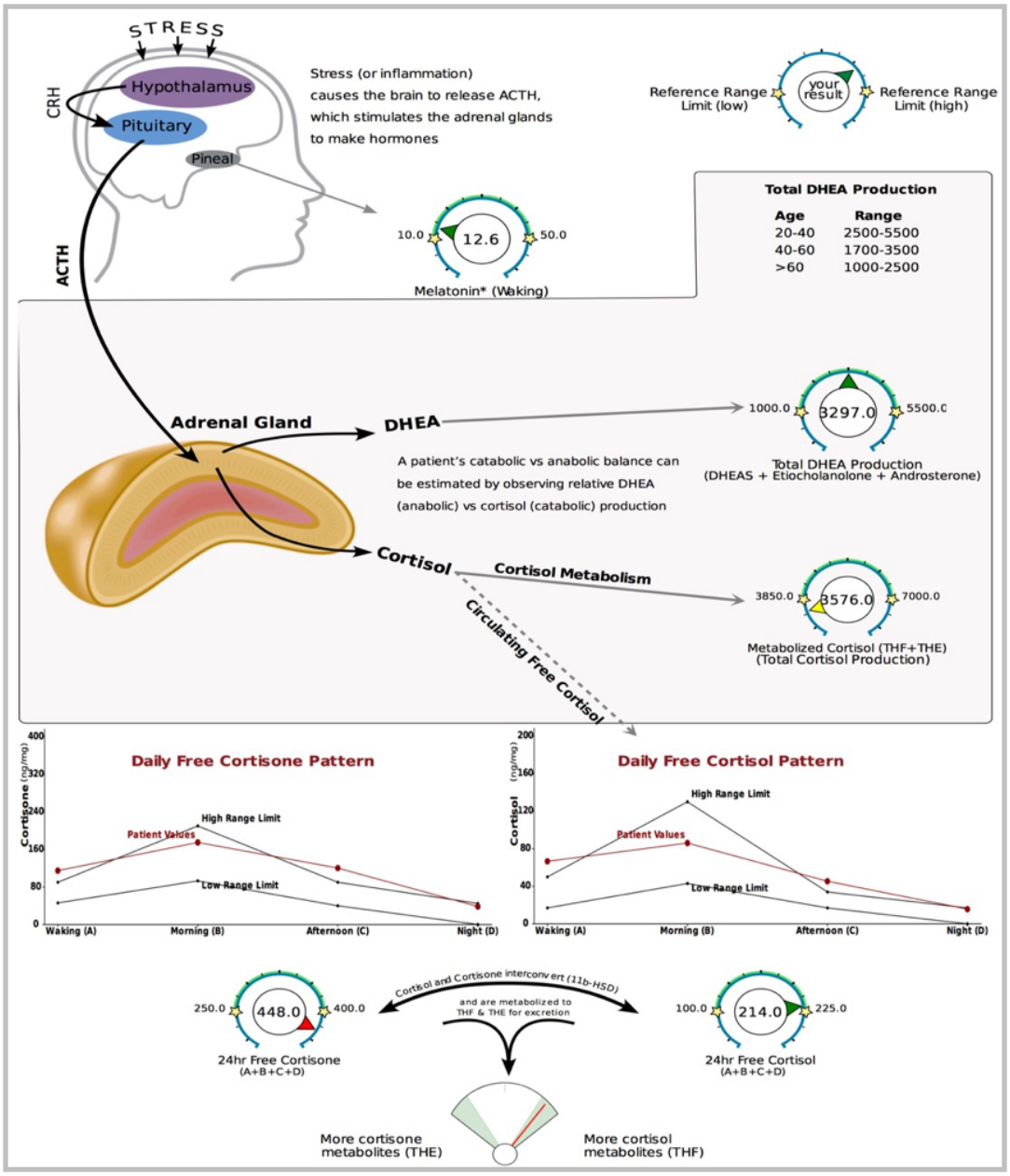
This study showed that patients with chronic fatigue syndrome have low free cortisol and low free cortisone in the urine compared with controls, so if you only tested free cortisol, that would kind of support the adrenal fatigue idea, but when you look at the cortisol metabolites, there was no difference in cortisol metabolites between the chronic fatigue syndrome patients and the control. So in this case, the chronic fatigue patients had low free cortisol and free cortisone but normal cortisol metabolites.

The last pattern that we're going to discuss is the other discordant pattern with high free cortisol and low metabolized cortisol, and this is observed in hypothyroidism, licorice supplementation, inflammation, and in some cases, it may be normal and nonpathological.



A few slides back we talked about how hypothyroidism can present with normal or high free cortisol and low metabolites, and here is an example of this. This is a patient, a 52-year-old male, with no significant complaints. He just wanted to optimize his health. Free cortisol was high normal. Metabolized cortisol was low, and he had a TSH, a thyroid-stimulating hormone, of 3.2. Now we're going to cover this in the blood chemistry section, but 3.2 is within the reference range for TSH, but it's higher than what it should be, and I'm going to explain why the reference range is wrong in the case of TSH. TSH for a normally functioning thyroid should be 2.5 or 2.2 or below, depending on the studies that you look at.

So his labs were indicating some thyroid dysfunction, and this is showing up here in the urine results, and this is one of the reasons why researchers are considering using urinary cortisol metabolites or cortisol test results as a way of diagnosing subclinical hypothyroidism.



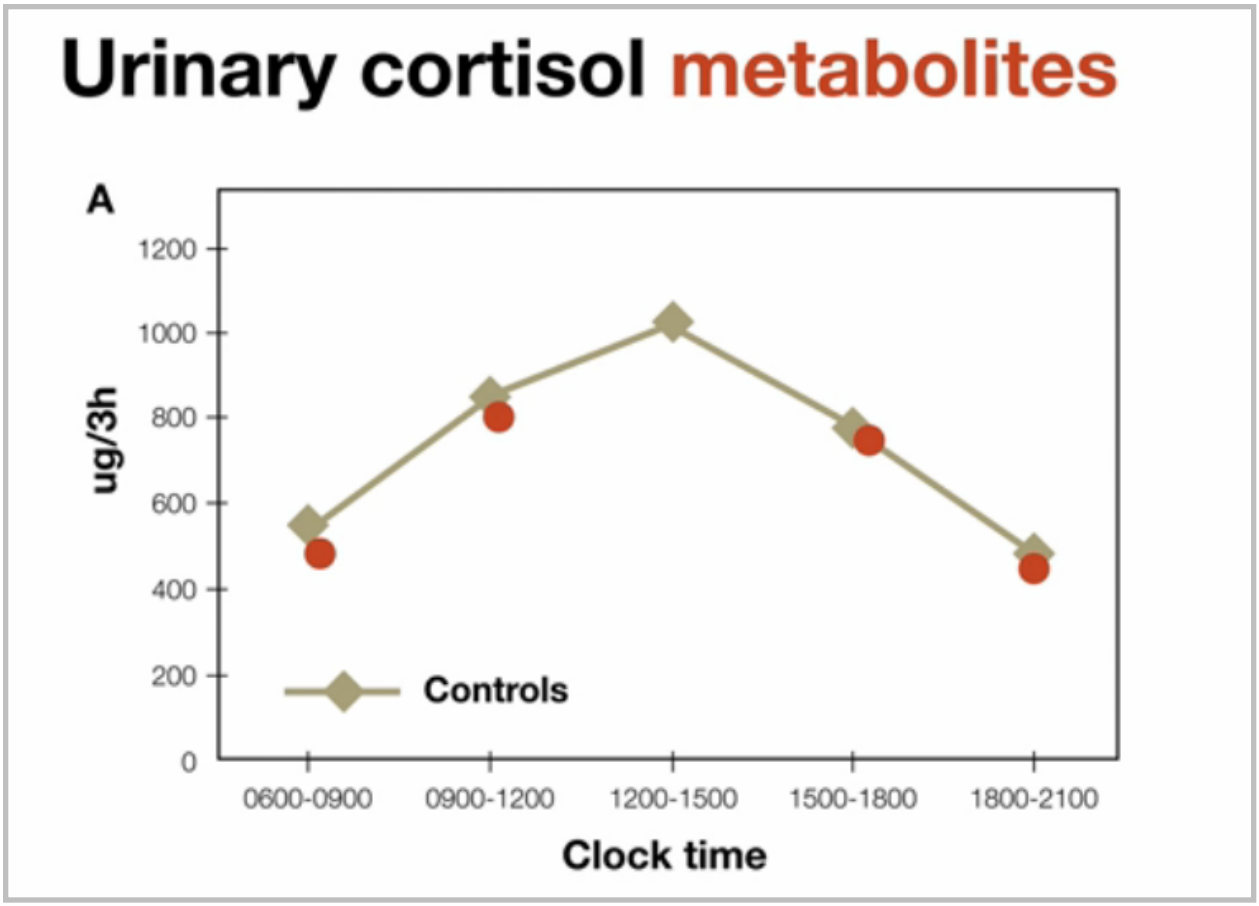
Here's another example. This is a 38-year-old male with chief complaints of constipation and fatigue, which are, of course, classic hypothyroid symptoms. His TSH was 4.35. He had a history of thyroid disease in the family and high C-reactive protein. His free T4, free T3 levels, and thyroid antibodies were normal, but symptoms, his TSH, and this test clearly are showing some thyroid issue.

Licorice contains a compound called glycyrrhizin, which has a structure that is very similar to corticosteroids. Metabolites of glycyrrhizin have been shown to block 11β-HSD2, which is the enzyme responsible for the conversion of cortisol to cortisone. This causes a buildup of free cortisol, especially in tissues attempting to block cortisol binding to mineralocorticoid receptors like the kidney. Chronic high intake of licorice has been shown to raise blood pressure through cortisol binding of mineralocorticoid receptors in the kidney, which leads to pseudoaldosteronism, increasing water retention and blood volume. This only happens, however, with people who already had normal or elevated cortisol, and it's not typically a concern in people with low cortisol, but again, it highlights the importance of measuring total cortisol and not just free cortisol levels.

Inflammation and cortisol/DHEA

Free cortisol	High
Total metabolites	Normal or low
Cortisol to cortisone ratio	Increased
DHEA (S)	Normal
DHEA-Sulfate	Low

Inflammation can also cause high free cortisol with normal or low metabolites. Inflammation pushes cortisone back to cortisol via 11β-HSD1, so I'm going to talk about this a little more later in the presentation, but DHEA sulfate is often low in inflammatory conditions because inflammation inhibits sulfation. So DHEA metabolites may be normal, and DHEA overall isn't low, just DHEA sulfate is low because of inflammation. So I put a table here on the slide with some of the findings that you could expect to see with inflammation. You might see high free cortisol. You would see maybe normal or low cortisol metabolites. You'd see an increased cortisol-to-cortisone ratio. You might see normal total DHEA, but DHEA sulfate would be low.



Finally, in some cases, you'll see high free cortisol with normal or low metabolized cortisol that is not pathological. When you see this pattern, ask yourself this question: Which of the free cortisol readings is the highest? Cortisol obviously maxes out in the early morning sample because overnight and early morning are when cortisol production is the highest, but the metabolites lag behind by about 90 minutes when you're using the dried urine test, remember. So in the figure on the slide, samples were collected at the times where you see the orange dots. So the absolute max that comes out in the early afternoon corresponding to the early morning cortisol is missed, so if someone is relatively high in the early morning and lower the rest of the day, the DUTCH test will miss a disproportionate amount of the metabolites that come from that morning surge, and that is one disadvantage of the 90-minute lag time that is inherent to urine samples. So if you see this pattern where someone is relatively high in free cortisol in the early morning and lower the rest of the day, you might see metabolites that are lower because they'll miss that disproportionate amount in the morning, and that is not necessarily a pathological finding, so keep the timing in mind when you're reviewing the results.

Okay, that's it for part 1. In part 2, we'll cover some additional test results and patterns. I'll talk to you then.