Urinary Hormone Assessment

Why Test Hormones in the Urine?

**Provides an average of fluctuating hormones**
Steroid hormone levels naturally fluctuate over the course of a day. Measurement of hormones in urine collected over several hours provides a stable average of hormone levels that overcomes the “snapshot” limitation inherent in serum and salivary testing.¹

**Evaluates hormone utilization**
Steroid hormones that are produced primarily in the adrenal cortex, ovaries and testes are extensively metabolized in various tissues. How they are metabolized influences the ultimate clinical effect of these hormones; this can vary significantly. Measuring their downstream metabolites in the urine can provide us with this critical intracellular information.²

**Identifies bioavailable fraction of hormones**
The majority of steroid hormones are bound to proteins in the blood that help to maintain these hormones in a non-active ‘reserve’ form. It is only the tiny, unbound fraction that is available to act upon the body via cellular hormone receptors. The concentration of binding proteins can vary, making it impossible to predict the bioavailable fraction of hormone from a measurement of total (bound and unbound) hormone. All hormones measured in urine represent the unbound, or bioavailable, fraction.

Are there any limitations to urinary hormone testing?

**Abnormal Renal Function**
Assessment of any compounds in the urine depends upon normal kidney function. Therefore, hormones in patients with abnormal renal function should be evaluated via blood or saliva.

**Evaluation of Diurnal Rhythms**
Even though averaging hormonal fluctuations in urine offers an advantage, there are still times when evaluating diurnal rhythms of certain hormones via saliva can provide valuable information. An example is cortisol, which normally shows a robust output after waking, with a gradual decline over the remainder of the day. Salivary cortisol testing (comprising four timed salivary collections over the course of the day) can help reveal uncharacteristic spikes or drops in cortisol at certain points in the day, or a flattening out of the diurnal rhythm, a sign of stress-induced maladaptation and dysfunction within the hypothalamic/pituitary/adrenal (HPA) axis.

**Hematuria**
Finally, blood in the urine may compromise the accuracy of urinary steroid results, mostly due to the hormonal content of red blood cells contributing to the total measurement.
How do I select the most appropriate profile for a patient?

**Adrenal steroids**

Adrenal steroid evaluation might be considered for patients with dysglycemia (e.g., metabolic syndrome), tissue wasting disorders or low bone density, chronic fatigue, immune dysfunction, hypo- or hypertension, allergies, menstrual irregularities, or simply a history of chronic psychological or physiologic stress.

Adrenal assessment is also useful in hypothyroid patients. CRH, which is released from the hypothalamus during stress but also in response to low cortisol, inhibits TSH secretion and thyroid function. High amounts of glucocorticoids also inhibit TSH secretion. Glucocorticoids and adrenaline render target tissues less sensitive to the effects of thyroid hormone, as well as inhibit the conversion of T4 to T3. In light of these effects, addressing adrenal imbalances before administering thyroxine makes sense, and may help improve tolerance of thyroxine in those patients who require it.3

**Androgens**

Androgen evaluation should be considered for patients experiencing decreased libido or sense of well-being, bone loss, signs and symptoms of “andropause” in men (e.g., loss of muscle mass, increased adipose, decreased drive), or signs of hyperandrogenism (e.g., hirsutism, alopecia, acne) or history of PCOS in women.

**Estrogens and progesterone**

Estrogens and pregnanediol (metabolite of progesterone) levels can help evaluate ovulatory function in pre-menopausal women with menstrual irregularities and/or infertility, as well as menopausal symptoms in peri- or post-menopausal women. Estrogen metabolites help assess the risk of breast or prostate cancer, osteoporosis (in postmenopausal women not using HRT), and autoimmune conditions such as lupus and rheumatoid arthritis.4 These markers are also helpful in monitoring patient response to interventions such as indole-3-carbinol (I3C), diindolylmethane (DIM), and sulforaphane.

**Comprehensive analysis**

Given the close interrelationships of steroid hormones, a broad-spectrum assessment comprising all of these metabolites is recommended, at least for an initial evaluation. Smaller sub-panels may be employed for follow-up testing to monitor response to treatments or/to check levels of previously abnormal results. As mentioned above, collections may be 24 hour collections or first-morning void.
Breast & Prostate Cancer Risk Assessment via Estrogen Metabolites

It is not just the amount of total circulating estrogen that impacts breast and prostate cancer risk, but also how that estrogen is processed. Estrogen can be metabolized in healthy, protective directions, or in unhealthy directions that increase cancer risk. Urine testing provides the most comprehensive evaluation of estrogen metabolism possible.

Healthy Phase 1 metabolism: The enzyme CYP1A1 converts estrone to 2-hydroxyestrone (2-OHE1).

Unhealthy Phase 1 metabolism: CYP3A4 and CYP1B1 convert estrone to 16α-hydroxyestrone (16α-OHE1) and 4-hydroxyestrone (4-OHE1), respectively.

Healthy Phase 2 metabolism: COMT (methylation enzyme) neutralizes 4-OHE1 to 4-methoxyestrone (4-MeOE1). COMT also converts 2-OHE1 to its most protective form, 2-methoxyestrone (2-MeOE1).²

LOWER RISK of breast or prostate cancer is associated with:
- Higher 2-OHE1
- Higher 2-OHE1/16α-OHE1 ratio
- Higher 2-MeOE1/2-OHE1 ratio
- Higher 4-MeOE1/4-OHE1 ratio

HIGHER RISK of breast or prostate cancer is associated with:
- Higher 16α-OHE1
- Higher 4-OHE1
- Lower 2-OHE1/16α-OHE1 ratio³⁻⁴(also associated with RA and lupus)⁶⁻¹¹
- Lower 2-MeOE1/2-OHE1 ratio
- Lower 4-MeOE1/4-OHE1 ratio
**TREATMENT OPTIONS to improve estrogen metabolism:**

**Lower 2-OHE1 and/or 2-OHE1/16α-OHE1 ratio, or higher 4-OHE1?**
- Increase intake of cruciferous vegetables.
- Consider broccoli derivatives such as:
  - indole-3-carbinol (I3C) – (150 mg 2-3 X day)
  - diindolylmethane (DIM) – (120 mg 1-2 X day)
  - sulforaphane – (30 mg/day)
- Rule out hypothyroidism, toxicity (e.g., pesticides, polycyclic aromatic hydrocarbons, PCBs).

**Lower 2-MeOE1/2-OHE1 and/or 4-MeOE1/4-OHE1 ratios?**
- Provide nutritional support for methylation:
  - Ensure adequate protein and/or assimilation of protein
  - Ensure adequate magnesium, B2, B6, B12, folic acid, serine
  - Consider betaine (trimethylglycine, or TMG) – (500 mg 2-3 X day)

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**HRT and Hormone Balance Assessment**

Bio-identical hormones are fully represented in a urinary hormone evaluation, including their downstream metabolites. Because urinary hormones offer the best reflection of how the body is processing exogenous hormones, steroid measurement in the urine helps ensure that the therapy is having the desired effect.

**NOTE:** Oral contraceptives and other synthetic hormones, such as medroxyprogesterone acetate (e.g., Provera®) are not reflected in any laboratory assay, including urine. Conjugated equine estrogens (e.g., Premarin®) are only partially represented, although the balance of their estrone metabolites may help shed light on breast cancer risk.

**NOTE:** Urinary hormones are available as 24-hour collections or First-Morning Void (FMV), which reflects nocturnal levels of hormones. Because of the variability of HRT delivery systems and their pharmacokinetics, it may be easier to obtain a reliable ‘average’ by using the 24-hour collection rather than a First-Morning Void in HRT-supplemented individuals.

**ESTROGENS** (estradiol, estrone, estriol, and estrogen metabolites) in the urine reflect circulating levels as well as tissue metabolism. As a result of metabolism, serum estradiol is largely represented as estrone (glucuronide) in the urine. Estrogen metabolism is discussed above.

<table>
<thead>
<tr>
<th>Hormone</th>
<th>Reference Range</th>
</tr>
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<tbody>
<tr>
<td>Estrone</td>
<td></td>
</tr>
<tr>
<td>Follicular</td>
<td>3.60-57.60 mcg/24 hr</td>
</tr>
<tr>
<td>Midcycle</td>
<td>24.00-156.00 mcg/24 hr</td>
</tr>
<tr>
<td>Luteal</td>
<td>10.80-72.00 mcg/24 hr</td>
</tr>
<tr>
<td>Menopausal</td>
<td>0.30-7.90 mcg/24 hr</td>
</tr>
<tr>
<td>Male</td>
<td>3.00-28.50 mcg/24 hr</td>
</tr>
</tbody>
</table>

Reference ranges are gender-specific and related to either menstrual status (24 hr) or age (FMV).
**PROGESTERONE** is metabolized into numerous downstream compounds, but most immediately into pregnanediol and pregnanetriol. Urine levels of these metabolites tend to correlate with levels of serum progesterone, thus may be used to assess status of the hormone.13

<table>
<thead>
<tr>
<th>Metabolite</th>
<th>Level (24hr urine)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pregnanediol (24hr urine)</td>
<td>0.31</td>
</tr>
<tr>
<td>Pregnanetriol (24hr urine)</td>
<td>0.51</td>
</tr>
</tbody>
</table>

**ANDROGENS** such as DHEA, testosterone, and androstenedione tend to be extensively metabolized into downstream androgen compounds. As a result, the levels of testosterone along with “Total 17-ketosteroids” (DHEA plus metabolites) should be used to assess androgen status and/or efficacy of androgen replacement.

<table>
<thead>
<tr>
<th>Metabolite</th>
<th>Level (24hr urine)</th>
</tr>
</thead>
<tbody>
<tr>
<td>17-Ketosteroids, Total* (24hr urine)</td>
<td>10.51</td>
</tr>
</tbody>
</table>

**Total 17-ketosteroids include the following:**

<table>
<thead>
<tr>
<th>Metabolite</th>
<th>Level (24hr urine)</th>
</tr>
</thead>
<tbody>
<tr>
<td>DHEA (24hr urine)</td>
<td>&lt;di</td>
</tr>
<tr>
<td>Androsterone (24hr urine)</td>
<td>2.56</td>
</tr>
<tr>
<td>Etiocholanolone (24hr urine)</td>
<td>2.38</td>
</tr>
<tr>
<td>11-Keto-androsterone (24hr urine)</td>
<td>&lt;di</td>
</tr>
<tr>
<td>11-Keto-etiocholanolone (24hr urine)</td>
<td>0.70</td>
</tr>
<tr>
<td>11-Hydroxy-androsterone (24hr urine)</td>
<td>3.36</td>
</tr>
<tr>
<td>11-Hydroxy-etiocholanolone (24hr urine)</td>
<td>1.40</td>
</tr>
</tbody>
</table>

**Lower level of Total 17-Ketosteroids may be associated with:**
- Acute or chronic stress (check Total 17-Hydroxy-corticosteroids)
- Aging, DHEA deficiency
- Excessive conversion of androgens to estrogens (see “Aromatase activity” below)
- High estrogen level or estrogen replacement including oral contraceptives (can increase sex-hormone binding globulin, which reduces the amount of bioavailable androgens)
- Smoking, chronic alcohol ingestion
- Diabetes mellitus
- Ketoconazole, metformin, troglitazone

**TREATMENT OPTIONS to increase Total 17-Ketosteroids:**
- Stress management (see section for Total 17-Hydroxy-corticosteroids)
- Increase dietary protein.
- Address aromatase imbalances.
- Consider supplementation with DHEA (15-100 mg/day) or other androgens.

**Higher level of Total 17-Ketosteroids may be associated with:**
- DHEA, androstenedione, or testosterone supplementation
- High protein- or high calorie diet
- PCOS (especially with obesity) and/or hirsutism and/or acne
- Low sex-hormone binding globulin (e.g., from hyperinsulinemia or hypothyroidism)
- Aromatase inhibitors
- Congenital adrenal hyperplasia
- Tumors of adrenals, ovaries, or testes

**TREATMENT OPTIONS to decrease Total 17-Ketosteroids:**
- Improve insulin sensitivity.
- Correct thyroid imbalances.
AROMATASE ACTIVITY:
The enzyme aromatase converts androgens to estrogens. *If enzyme activity is excessive, then administration of testosterone or DHEA may mostly serve to increase levels of estrogen.*

HIGHER aromatase activity is suggested by:
- Lower Total 17-ketosteroids along with higher estrogens

**TREATMENT OPTIONS to reduce aromatase:**
- Weight loss (greater aromatase activity in adipose)
- Reduce stress and/or inflammation.
- Improve insulin sensitivity.
- Natural aromatase inhibitors:
  - Chrysin (1-3 gms/day, oral; 100-300 mg/day transdermal))
  - Green tea and/or green tea extract (500 mg 1-3 X day)
  - Epilobium (250 mg 1-2 X day)
  - Flaxseed (5-10 gms/day)
  - Stinging nettles (300 mg 1-2 X day, freeze-dried leaf)
  - Vitamin C (500 mg 1-2 X day)
- Pharmaceutical aromatase inhibitors:
  - Anastrozole (Arimidex®) (1 mg/day)
  - Exemestane (Aromasin®) (25 mg/day)
  - Letrozole (Femara®) (2.5 mg/day)

5-ALPHA-REDUCTASE ACTIVITY:
The enzyme 5-alpha reductase converts testosterone to dihydrotestosterone (DHT), a potent androgen associated with male-pattern baldness, acne, hirsutism, polycystic ovarian syndrome, prostate hypertrophy and cancer risk. Alternatively, testosterone can be metabolized by 5-beta reductase to an inactive metabolite.

Possible indicators of higher 5α-reductase activity include:
- Higher androstanediol
- Higher androstenedione
- Lower etiocholanolone
- Lower E:A (5α:5α) ratio
- THF:a-THF ratio

**TREATMENT OPTIONS to reduce 5α-reductase:**
- Reduce carbohydrate intake.
- Herbal 5α reductase inhibitors:
  - Saw palmetto (160 mg 2 X day, extract)
  - Pygeum (50-150 mg/day)
  - Stinging nettle nettles (300 mg 1-2 X day, freeze-dried leaf)
- Flaxseed (5-10 gms/day)
- Soy isoflavones (20-80 mg day)
- Green tea and/or green tea extract (500 mg 1-3 X day)
- Quercetin (200-400 mg 1-3 X day)
- Progesterone
- Pharmaceutical 5α-reductase inhibitors:
  - Finasteride (Proscar® – 5 mg/day; Propecia® – 1 mg/day)
  - Dutasteride (Avodart®) (0.5 mg/day)
Adrenal Stress Assessment & Anabolic/Catabolic Balance

Adrenal Stress:
Stress increases cortisol production, which can be life-saving in the short term. Chronic stress, however, can lead to cortisol-induced immune suppression, hyperglycemia, insulin resistance, central obesity, hypertension, memory impairment, hyperlipidemia, and/or altered thyroid function. Low cortisol production and/or HPA axis impairment, including low diurnal variation in cortisol secretion, may follow chronic over-activation of the axis by chronic stress, leading to fatigue and other problems. 

Greater than 95% of cortisol is metabolized before excretion. Urinary “Total hydroxy-corticosteroids” (17-OHCS) account for more than 50% of cortisol’s metabolic byproducts, thus can be used along with cortisol to help gauge overall glucocorticoid production and metabolism.

Cortisol:

<table>
<thead>
<tr>
<th>Test</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cortisol, Free (24hr urine)*</td>
<td>60</td>
</tr>
<tr>
<td>17-Hydroxy-corticosteroids, Total* (24hr urine)</td>
<td>38.58</td>
</tr>
</tbody>
</table>

Total 17-hydroxy-corticosteroids include the following:

- Pregnanetriol (24hr urine) | 2.38 |
- allo-Tetrahydrocortisol, a-THF (24hr urine) | 9.89 |
- Tetrahydrodeoxycortisol (24hr urine) | 0.64 |
- Tetrahydrocortisone, THE (24hr urine) | 22.33 |
- Tetrahydrocortisol, THF (24hr urine) | 9.66 |

NOTE: Cortisol is not included in the Total 17-OHCS for two reasons: 1) Cortisol and the 17-OHCS are prepared and measured differently; and 2) all research examining 17-OHCS has utilized the GC-MS method only, thus excluding cortisol.

Lower urinary cortisol along with lower Total 17-hydroxy-corticosteroids suggests:
- Low cortisol production, as in adrenal insufficiency (e.g., chronic stress, fibromyalgia, chronic fatigue syndrome, or Addison’s disease) or congenital adrenal hyperplasia (metabolic enzyme deficiency) or
- Suppression of endogenous cortisol production from exogenous glucocorticoids such as Prednisone* or
- Pituitary insufficiency

Higher urinary cortisol along with higher Total 17-hydroxy-corticosteroids suggests:
- High cortisol production (e.g., severe stress, strenuous exercise, anorexia nervosa, or Cushing’s disease) or
- Normal or low cortisol production, along with increased clearance and metabolism of cortisol (e.g., obesity* or chronic stress)*

*NOTE: Urinary cortisol reflects both the production and the excretion of cortisol. Therefore, it is highly recommended to also check serum a.m. cortisol or timed salivary collections throughout the day to help identify patients with low baseline levels and/or impaired HPA axis function (flattened curve).
TREATMENT OPTIONS for adrenal excess (confirmed by elevated salivary or serum cortisol):

- Stress management (adequate sleep, meditation, yoga, or other stress-reducing activities; avoidance of overscheduling and negative thinking, etc; breathe, move, laugh)
- Minimize stimulants such as caffeine, ephedra, yerba mate.
- Identify and address sources of physiologic stress (e.g., inflammatory conditions, allergies, reactive hypoglycemia, dysbiosis/leaky gut, or infection).
- Address causes of persistent cortisol activation or production:
  - Excess adipose, especially abdominal / high insulin / metabolic syndrome
  - Hypothyroidism
  - Inflammation
- Adequate intake of vitamin C, magnesium, zinc, B vitamins; consider additional B6 (100 mg 1-2 X day) and pantothenic acid (500 mg 2-3 X day)
- Phosphatidylserine to calm HPA axis activation and prevent neuronal cortisol toxicity (300-800 mg/day)
- Calmative herbal agents such as passiflora, hypericum, valerian, scutellaria, magnolia bark, hops (consult companies for product dosing recommendations)
- Calmative nutritional agents such as GABA (250 mg 1-3 X day), L-theanine (100 mg 3-4 X day)
- Herbal “adaptogenic” herbs such as eleutherococcus (Siberian ginseng), ashwagandha, astragalus, schizandra, rhodiola (consult companies for product dosing recommendations)
- Rule out licorice ingestion (prevents cortisol breakdown).  
- DHEA to balance cortisol (check Total 17-ketosteroids) and/or plant sterols/sterolins (reduces cortisol, balances cortisol/DHEA ratio).

TREATMENT OPTIONS for adrenal insufficiency (confirmed by low salivary or serum cortisol):

- Stress management (adequate sleep, meditation, yoga, or other stress-reducing activities; minimize over-scheduling, negative thinking, etc.)
- Address sources of chronic physiologic stress (e.g., inflammatory conditions, allergies, reactive hypoglycemia, dysbiosis/leaky gut, infection).
- Gentle exercise
- Minimize intake of sweets and simple carbohydrates.
- Adequate intake of vitamin C, magnesium, zinc, B vitamins; consider additional B6 (100 mg 1-2 X day) and pantothenic acid (500 mg 2-3 X day)
- Herbal “adaptogenic” herbs such as eleutherococcus (Siberian ginseng), Panax ginseng, ashwagandha, astragalus, schizandra, rhodiola, licorice (consult companies for product dosing recommendations)
- Bovine glandular supplements (e.g., adrenal, hypothalamus, pituitary), adrenal cortical extract, or low-dose hydrocortisone (e.g., 5 mg 4 X day), only after low cortisol has been confirmed by salivary and/or serum cortisol; always balance with DHEA) (Consult companies for product dosing recommendations.)

Anabolic/Catabolic Balance

Persistent catabolic effects of cortisol are most damaging when not balanced by anabolic hormones such as dehydroepiandrosterone (DHEA), important in functions such as immunity, reproduction (via its conversion to downstream sex steroids), cardiovascular health, bone density, and memory.

This anabolic/catabolic balance – or the balance of ‘growth and healing’ versus ‘wear and tear’ in the body – can be assessed by comparing total 17-hydroxycorticosteroids with total 17-ketosteroids in the urine.
A low Anabolic/Catabolic balance may be associated with:

- A metabolic shift in which the production of stress hormone is favored over the production of androgens and downstream estrogens
- Aging
- Deficient growth hormone
- Chronic stress or glucocorticoid therapy
- Excessive exercise
- Acute or chronic illness, reduced ability to recover from illness or injury
- Increased risk of developing disease, poorer prognosis in patients with illness\(^{27,28}\)

**TREATMENT OPTIONS to increase Anabolic/Catabolic Balance:**

- Refer to appropriate Treatment Options for adrenal excess (high Total 17-hydroxy-corticosteroids) or low Total 17-ketosteroids, depending on which imbalance is contributing to the low ratio.

A high Anabolic/Catabolic balance may be associated with:

- Excessive androgen therapy or a metabolic shift in which androgen production is favored over cortisol production
- Possible polycystic ovarian syndrome and/or hirsutism
- Congenital adrenal hyperplasia (enzyme deficiencies)
- Adrenal tumors
- Extreme athleticism

**TREATMENT OPTIONS to decrease Anabolic/Catabolic Balance:**

- Refer to appropriate Treatment Options for adrenal insufficiency (low Total 17-hydroxy-corticosteroids) or high Total 17-ketosteroids, depending on which imbalance is contributing to the high ratio.
References


