Overview

Useful For
Screening for Cushing syndrome
Diagnosis of Cushing syndrome in patients presenting with symptoms or signs suggestive of the disease

Special Instructions
- Cortisol - Saliva Collection Instructions

Method Name
Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS)

NY State Available
Yes

Specimen

Specimen Type
Saliva

Necessary Information
Collection time is required.

Specimen Required
Patient Preparation:
1. Do not brush teeth before collecting specimen.
2. Do not eat or drink for 15 minutes prior to specimen collection.

Supplies: Cortisol, Saliva Collection Kit (T514)

Container/Tube: SARSTEDT Salivette

Specimen Volume: 1.5 mL

Collection Instructions:
1. Provide patient with a Saliva Collection Kit (Salivette) containing the Cortisol - Saliva Collection Instructions and ask them to follow the instructions as written.
2. Instruct patient to collect specimen between 11 p.m. and midnight and record collection time on the Cortisol - Saliva Collection Instructions sheet.
3. Instruct patient to return Cortisol - Saliva Collection Instructions with the appropriately labeled Salivette to the laboratory.

Additional Information:
1. Reference values are also available for an 8 a.m. (7 a.m.-9 a.m.) or a 4 p.m. (3 p.m.-5 p.m.) collection, however, the 11 p.m. to midnight collection is preferred.

2. If multiple specimens are collected, submit each vial under a separate order.

**Specimen Minimum Volume**

0.6 mL

**Reject Due To**

All specimens will be evaluated at Mayo Clinic Laboratories for test suitability.

**Specimen Stability Information**

<table>
<thead>
<tr>
<th>Specimen Type</th>
<th>Temperature</th>
<th>Time</th>
<th>Special Container</th>
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</thead>
<tbody>
<tr>
<td>Saliva</td>
<td>Refrigerated (preferred)</td>
<td>28 days</td>
<td></td>
</tr>
<tr>
<td></td>
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<tr>
<td></td>
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**Clinical and Interpretive**

**Clinical Information**

Cortisol levels are regulated by adrenocorticotropic hormone (ACTH), which is synthesized by the pituitary in response to corticotropin-releasing hormone (CRH). Cushing syndrome results from overproduction of glucocorticoids because of either primary adrenal disease (adenoma, carcinoma, or nodular hyperplasia) or an excess of ACTH (from a pituitary tumor or an ectopic source). ACTH-dependent Cushing syndrome due to a pituitary corticotroph adenoma is the most frequently diagnosed subtype; most commonly seen in women in the third through fifth decades of life.

CRH is released in a cyclic fashion by the hypothalamus, resulting in diurnal peaks (elevated in the morning) and nadirs (low in the evening) for plasma ACTH and cortisol levels. The diurnal variation is lost in patients with Cushing syndrome and these patients have elevated levels of evening plasma cortisol. The measurement of late-night salivary cortisol is an effective and convenient screening test for Cushing syndrome. In a recent study from the National Institute of Health, nighttime salivary cortisol measurement was superior to plasma and urine free cortisol assessments in detecting patients with mild Cushing syndrome. The sensitivity of nighttime salivary cortisol measurements remained superior to all other measures. The distinction between Cushing syndrome and pseudo-Cushing states is most difficult in the setting of mild-to-moderate hypercortisolism. Subtle increases in salivary cortisol collected at midnight (cortisol of nadir) appear to be one of the earliest abnormalities in Cushing syndrome.

**Reference Values**

7 a.m.-9 a.m.: 100-750 ng/dL

3 p.m.-5 p.m.: <401 ng/dL

11 p.m.-midnight: <100 ng/dL

**Interpretation**

Cushing syndrome is characterized by increased salivary cortisol levels, and late-night saliva cortisol measurements may be the optimum test for the diagnosis of Cushing syndrome. It is standard practice to confirm elevated results at
least once. This can be done by repeat late-night salivary cortisol measurements, midnight blood sampling for cortisol (CORT / Cortisol, Serum), 24-hour urinary free cortisol collection (CORTU / Cortisol, Free, 24 Hour, Urine), or overnight dexamethasone suppression testing. Upon confirmation of the diagnosis, the cause of hypercortisolism, adrenal versus pituitary versus ectopic adrenocorticotropic hormone production, needs to be established. This is typically a complex undertaking, requiring dynamic testing of the pituitary adrenal axis and imaging procedures. Referral to specialized centers or in-depth consultation with experts is strongly recommended.

Cautions
Acute stress (including hospitalization and surgery), alcoholism, depression, and many drugs (eg, exogenous glucocorticoids, anticonvulsants) can obliterate normal diurnal variation, affect response to suppression/stimulation tests, and cause elevated cortisol levels.

Cortisol levels may be increased in pregnancy and with exogenous estrogens.

Midnight salivary cortisol assay cannot diagnose hypocortisolism or Addison disease because of the limited sensitivity of the assay method.

Supportive Data
Using this assay, it was determined that late-night salivary cortisol is in the range of 100 ng/mL to 6,000 ng/dL (2.76-166 nmol/L) for clinically confirmed Cushing patients (N=11).

Normal values are based on 36 donors (ages 0-8 years), 46 donors (ages 9-17 years), and 102 donors (age >17 years).

Clinical Reference

Performance
Method Description
Deuterated cortisol (d3-cortisol) is added to 0.1 mL sample as an internal standard. Cortisol and d3-cortisol are extracted from the specimen using online turbulent flow high-performance liquid chromatography (HPLC) and analyzed by liquid chromatography-tandem mass spectrometry (LC-MS/MS) using multiple reaction monitoring in positive mode. The following transitions are used for analysis: Cortisol: 363.3/121.1; d3-cortisol: 366.3/121.1. (Taylor RL, Machacek DA, Singh RJ: Validation of a high-throughput liquid chromatography-tandem mass spectrometry method for urinary cortisol and cortisone. Clin Chem 2002; 48:1511-1519)

PDF Report
No

Day(s) and Time(s) Test Performed
Monday, Wednesday, Friday; 4 p.m.

Analytic Time
2 days

Maximum Laboratory Time
Test Definition: SALCT
Cortisol, Saliva

4 days

Specimen Retention Time
2 weeks

Performing Laboratory Location
Rochester

Fees and Codes

Fees
- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their Regional Manager. For assistance, contact Customer Service.

Test Classification
This test was developed and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the U.S. Food and Drug Administration.

CPT Code Information
82533

LOINC® Information

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