Overview

Useful For
Second-tier testing of newborns with abnormal screening result for congenital adrenal hyperplasia

Genetics Test Information
Second-tier newborn screen for congenital adrenal hyperplasia

Special Instructions
- Blood Spot Collection Card-Spanish Instructions
- Blood Spot Collection Card-Chinese Instructions
- Blood Spot Collection Instructions

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

NY State Available
Yes

Specimen

Specimen Type
Whole blood

Necessary Information
Birth weight, time of birth, and gestational age are required.

Specimen Required
Supplies: Card - Blood Spot Collection (Filter Paper) (T493)

Preferred: Blood Spot Collection Card (T493)

Acceptable: Local newborn screening card, Whatman 903 filter paper, Ahlstrom 226 filter paper, Munktell filter paper

Specimen Volume: 2 Blood spots

Collection Instructions:
1. Do not use device or capillary tube containing EDTA to collect specimen.
2. Do not expose specimen to heat or direct sunlight.
3. Do not stack wet specimens.
4. Keep specimen dry.

5. Let blood dry on the filter paper at ambient temperature in a horizontal position for a minimum of 3 hours.

**Additional Information:**

1. For collection instructions, see [Blood Spot Collection Instructions](#) in Special Instructions.

2. For collection instructions in Spanish, see [Blood Spot Collection Card-Spanish Instructions](#) (T777) in Special Instructions.

3. For collection instructions in Chinese, see [Blood Spot Collection Card-Chinese Instructions](#) (T800) in Special Instructions.

**Forms**

If not ordering electronically, complete, print, and send an [Inborn Errors of Metabolism Test Request](#) (T798) with the specimen.

**Specimen Minimum Volume**

Blood spot: 1

**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>
</tr>
</thead>
<tbody>
<tr>
<td>Whole blood</td>
<td>Ambient (preferred)</td>
<td>90 days</td>
<td>FILTER PAPER</td>
</tr>
<tr>
<td></td>
<td>Frozen</td>
<td>90 days</td>
<td>FILTER PAPER</td>
</tr>
<tr>
<td></td>
<td>Refrigerated</td>
<td>90 days</td>
<td>FILTER PAPER</td>
</tr>
</tbody>
</table>

**Clinical and Interpretable**

**Clinical Information**

Congenital adrenal hyperplasia (CAH) is a group of disorders caused by inherited defects in steroid biosynthesis, most commonly, 21-hydroxylase deficiency (approximately 90% of cases) and 11-beta hydroxylase deficiency (approximately 5% of cases). The overall incidence of CAH due to 21-hydroxylase deficiency is approximately 1 in 15,000 live births. Individuals with CAH may present with life-threatening salt-wasting crises in the newborn period and incorrect gender assignment of virilized females, which occurs due to in utero exposure to reduced glucocorticoids and mineralocorticoids and elevated 17-hydroxyprogesterone (17-OHP) and androgens. Hormone replacement therapy, when initiated early, results in a significant reduction in morbidity and mortality. Therefore, newborn screening for CAH is desirable and has been implemented in all 50 states.

Immunoaassays are typically used to quantify 17-OHP as a marker for CAH in the newborn screen setting. However, these immunoassays are hampered by cross-reactivity of the antibodies with other steroids, yielding a high rate of false-positive results. Tandem mass spectrometry (MS/MS) allows for the simultaneous specific determination of 17-OHP and other steroids, such as androstenedione, cortisol, 11-deoxycortisol, and 21-deoxycortisol. Application of
this technology to the determination of steroids in newborn blood spots significantly enhances the correct identification of patients with CAH and reduces the number of false-positive screening results when implemented as a second-tier analysis performed prior to reporting of initial newborn screen results.

**Reference Values**

**17-HYDROXYPROGESTERONE**

<15.1 ng/mL

**ANDROSTENEDIONE**

<3.1 ng/mL

**CORTISOL**

Not applicable

**11-DEOXYCORTISOL**

<15.1 ng/mL

**21-DEOXYCORTISOL**

<4.1 ng/mL

**(17 OHP + ANDROSTENEDIONE)/CORTISOL RATIO**

<1.1

**Note:** Abnormal (17 OHP + Androstenedione)/Cortisol Ratio: > or =1.1 is only applicable when 17-OHP is elevated

**11-DEOXYCORTISOL/CORTISOL RATIO**

Not applicable

**Interpretation**

Findings of a 17-hydroxyprogesterone (17-OHP) value greater than 15.0 ng/mL and a high (17-OHP + androstenedione)/cortisol ratio (> or =1) are supportive of the initial abnormal newborn screening result.

Findings of an 11-deoxycortisol value greater than 15.0 ng/mL or 21-deoxycortisol greater than 4.0 ng/mL with elevated 17-OHP further support the abnormal newborn screening result and increase the diagnostic specificity. Clinical and laboratory follow-up is strongly recommended.

**Cautions**

This is a screening test and, while it's positive predictive value is significantly higher than that of immunoassays (9.0% versus 0.5%), false-positive results can occur. Follow-up of abnormal results is necessary; perform OHPG / 17-Hydroxyprogesterone, Serum and DOC / 11-Deoxycortisol, Serum.

Deoxycorticosterone (DOC) coelutes with 17-OHP. DOC is a precursor of aldosterone in the steroidogenesis pathway. It is not elevated in patients with classic congenital adrenal hyperplasia (CAH) due to 21-alpha-hydroxylase deficiency, indeed its production is lacking in this deficiency. However, DOC is expected to be elevated in other variants of CAH, specifically 11-alpha-hydroxylase deficiency (which also includes elevations of 11-deoxy) and
17-alpha-hydroxylase/17,20-lyase deficiency (none of the other targeted steroids are elevated in this condition). Therefore, if this assay was applied as a first-tier screening assay, a finding of elevated DOC and/or 17-OHP can uncover clinically relevant variants of CAH that would be confirmed through follow-up testing. Because this assay’s goal is to identify patients with CAH but not exclusively 17-alpha-hydroxylase deficiency, the separation of 17-OHP and DOC during screening is not essential and may lead to a quicker diagnosis of at-risk patients for these more rare variants of CAH. However, as a second-tier test performed only when the first-tier test for 17-OHP is abnormal, 17-alpha-hydroxylase/17,20-lyase deficiency will not be uncovered because 17-OHP is not elevated in that condition.

Clinical Reference


Performance

Method Description

A 1/8-inch disk is punched out of the blood spot into a 96-well filter plate. Internal standards are added to the filter plate. The punched disks are eluted and centrifuged, then dried under nitrogen and reconstituted with 75:25 water:methanol. Analysis is by electrospray liquid chromatography-tandem mass spectrometry (LC-MS/MS). The concentration of 17-hydroxyprogesterone, androstenedione, cortisol, 11-deoxycortisol, and 21-deoxycortisol are established by comparison of their ion intensity to that of their respective internal standards.(Unpublished Mayo method)

PDF Report

No

Day(s) and Time(s) Test Performed

Monday through Saturday; 8 a.m.

Analytic Time

1 day (not reported on Sunday)

Maximum Laboratory Time

3 days

Specimen Retention Time

1 year

Performing Laboratory Location

Rochester

Fees and Codes
Test Definition: CAH2T
CAH Newborn Screen, BS

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
82542

LOINC® Information

<table>
<thead>
<tr>
<th>Test ID</th>
<th>Test Order Name</th>
<th>Order LOINC Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>CAH2T</td>
<td>CAH Newborn Screen, BS</td>
<td>57086-1</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Result ID</th>
<th>Test Result Name</th>
<th>Result LOINC Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>42207</td>
<td>17-OH Progesterone</td>
<td>38473-5</td>
</tr>
<tr>
<td>42208</td>
<td>Androstenedione</td>
<td>53343-0</td>
</tr>
<tr>
<td>42209</td>
<td>Cortisol</td>
<td>53345-5</td>
</tr>
<tr>
<td>42210</td>
<td>11-deoxycortisol</td>
<td>53338-0</td>
</tr>
<tr>
<td>42211</td>
<td>21-deoxycortisol</td>
<td>53341-4</td>
</tr>
<tr>
<td>42212</td>
<td>(17OHP+Androstenedione)/Cortisol</td>
<td>53336-4</td>
</tr>
<tr>
<td>42213</td>
<td>11-deoxycortisol/Cortisol</td>
<td>No LOINC Needed</td>
</tr>
<tr>
<td>BG688</td>
<td>Birth Weight (grams, XXXX)</td>
<td>8339-4</td>
</tr>
<tr>
<td>BG689</td>
<td>Time of Birth (24hr time, XX:XX)</td>
<td>57715-5</td>
</tr>
<tr>
<td>BG690</td>
<td>Gestational Age (weeks, XX.X)</td>
<td>76516-4</td>
</tr>
<tr>
<td>42206</td>
<td>Reviewed By</td>
<td>18771-6</td>
</tr>
<tr>
<td>42214</td>
<td>Interpretation (CAH2T)</td>
<td>46758-9</td>
</tr>
</tbody>
</table>