MAYO CLINIC LABORATORIES

Test Definition: 17PRN

Pregnenolone and 17-Hydroxypregnenolone, Serum

Overview

Useful For

An ancillary test for congenital adrenal hyperplasia (CAH), particularly in situations in which a diagnosis of both 21- and 11-hydroxylase deficiency have been ruled out

Confirming a diagnosis of 3-beta-hydroxysteroid dehydrogenase deficiency and 17-alpha-hydroxylase deficiency

As part of a battery of tests to evaluate women with hirsutism or infertility; both can result from adult-onset CAH

Profile Information

Test Id	Reporting Name	Available Separately	Always Performed
PREGN	Pregnenolone, S	Yes	Yes
17OHP	17-Hydroxypregnenolone, S	Yes	Yes

Testing Algorithm

For information see Steroid Pathways.

Special Instructions

- Steroid Pathways

Method Name

Liquid Chromatography Tandem Mass Spectrometry (LC-MS/MS)
Portions of this test are covered by patents held by Quest Diagnostics

NY State Available

Yes

Specimen

Specimen Type

Serum

Specimen Required

Supplies: Sarstedt Aliquot Tube, 5 mL (T914)
Collection Container/Tube:
Preferred: Red top

Acceptable: Serum gel
Submission Container/Tube: Plastic vial
Specimen Volume: 1 mL
Collection Instructions: Centrifuge and aliquot serum into a plastic vial.

Specimen Minimum Volume
0.5 mL

Reject Due To

Gross hemolysis	OK
Gross lipemia	OK
Gross icterus	OK

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Serum	Frozen	28 days	

Clinical & Interpretive

Clinical Information
Congenital adrenal hyperplasia (CAH) is caused by inherited defects in steroid biosynthesis. Deficiencies in several enzymes can cause CAH, including 21-hydroxylase (*CYP21A2* variants; 90% of cases), 11-hydroxylase (*CYP11A1* variants; 5%-8%), 3-beta-hydroxysteroid dehydrogenase (3-beta-HSD) (*HSD3B2* variants; <5%), and 17-alpha-hydroxylase (*CYP17A1* variants; <1%). The resulting hormone imbalances (reduced glucocorticoids and mineralocorticoids; and elevated steroid intermediates and androgens) can lead to life-threatening, salt-wasting crises in the newborn period and incorrect gender assignment of virilized females.

The adrenal glands, ovaries, testes, and placenta produce steroid intermediates, which are hydroxylated at position 21 (by 21-hydroxylase) and position 11 (by 11-hydroxylase) to produce cortisol. Deficiency of either 21-hydroxylase or 11-hydroxylase results in decreased cortisol synthesis and loss of feedback inhibition of adrenocorticotropic hormone (ACTH) secretion. The consequent increased pituitary release of ACTH drives increased production of steroid intermediates.

The steroid intermediates are oxidized at position 3 by 3-beta-HSD. The 3-beta-HSD enzyme allows formation of 17-hydroxyprogesterone (17-OHPG) from 17-hydroxypregnenolone and progesterone from pregnenolone. When 3-beta-HSD is deficient, cortisol is decreased, 17-hydroxypregnenolone and pregnenolone levels may increase, and 17-OHPG and progesterone levels, respectively, are low. Dehydroepiandrosterone is also converted to androstenedione by 3-beta-HSD and may be elevated in patients affected with 3-beta-HSD deficiency.

The best screening test for CAH, most often caused by either 21- or 11-hydroxylase deficiency, is the analysis of

17-OHPG (along with cortisol and androstenedione). CAH21 / Congenital Adrenal Hyperplasia (CAH) Profile for 21-Hydoxylase Deficiency, Serum allows the simultaneous determination of these 3 analytes. Alternately, these tests may be ordered individually: OHPG / 17-Hydroxyprogesterone, Serum; CINP / Cortisol, Mass Spectrometry, Serum; and ANST / Androstenedione, Serum.

If both 21- and 11-hydroxylase deficiency have been ruled out, analysis of 17-hydroxypregnenolone and pregnenolone may be used to confirm the diagnosis of 3-beta-HSD or 17-alpha-hydroxylase deficiency.

For more information see Steroid Pathways.

Reference Values

PREGNENOLONE
CHILDREN/ADOLESCENTS*
Males
0-6 years: Not established
7-9 years: <206 ng/dL
10-12 years: <152 ng/dL
13-15 years: 18-197 ng/dL
16-17 years: 17-228 ng/dL
Tanner Stages
Stage I: <157 ng/dL
Stage II: <144 ng/dL
Stage III: <215 ng/dL
Stage IV-V: 19-201 ng/dL

Females
0-6 years: Not established
7-9 years: <151 ng/dL
10-12 years: 19-220 ng/dL
13-15 years: 22-210 ng/dL
16-17 years: 22-229 ng/dL
Tanner Stages
Stage I: <172 ng/dL
Stage II: 22-229 ng/dL
Stage III: 34-215 ng/dL
Stage IV-V: 26-235 ng/dL

ADULTS
> or =18 years: 33-248 ng/dL

17-HYDROXYPREGNENOLONE
CHILDREN/ADOLESCENTS*
Males
Premature (26-28 weeks): 1,219-9,799 ng/dL

Premature (29-36 weeks): 346-8,911 ng/dL
Full term (1-5 months): 229-3,104 ng/dL
6 months-364 days: 221-1,981 ng/dL
1-2 years: 35-712 ng/dL
3-6 years: <277 ng/dL
7-9 years: <188 ng/dL
10-12 years: <393 ng/dL
13-15 years: 35-465 ng/dL
16-17 years: 32-478 ng/dL
Tanner Stages
Stage I: <209 ng/dL
Stage II: <356 ng/dL
Stage III: <451 ng/dL
Stage IV-V: 35-478 ng/dL

Females
Premature (26-28 weeks): 1,219-9,799 ng/dL
Premature (29-36 weeks): 346-8,911 ng/dL
Full term (1-5 months): 229-3,104 ng/dL
6 months-364 days: 221-1,981 ng/dL
1-2 years: 35-712 ng/dL
3-6 years: <277 ng/dL
7-9 years: <213 ng/dL
10-12 years: <399 ng/dL
13-15 years: <408 ng/dL
16-17 years: <424 ng/dL
Tanner Stages
Stage I: <236 ng/dL
Stage II: <368 ng/dL
Stage III: <431 ng/dL
Stage IV-V: <413 ng/dL

ADULTS
Males
> or =18 years: 55-455 ng/dL
Females
> or =18 years: 31-455 ng/dL

*Kushnir MM, Rockwood AL, Roberts WL, et al. Development and performance evaluation of a tandem mass spectrometry assay for 4 adrenal steroids. Clin Chem. 2006;52(8):1559-1567

To convert to nmol/L, multiply the value in ng/dL by 0.03159757.

Interpretation

The diagnosis and differential diagnosis of congenital adrenal hyperplasia (CAH) always require the measurement of several steroids. Patients with CAH due to steroid 21-hydroxylase gene (*CYP21A2*) variants usually have very high levels of androstenedione, often 5-fold to 10-fold elevations. 17-Hydroxyprogesterone (17-OHPG) levels are usually even higher, while cortisol levels are low or undetectable. All 3 analytes should be tested.

For the *HSD3B2* variant, cortisol, 17-OHPG, and progesterone levels will be decreased; 17-hydroxypregnenolone, pregnenolone, and DHEA levels will be increased.

In the much less common *CYP11A1* variant, androstenedione levels are elevated to a similar extent as seen in *CYP21A2* variant, and cortisol also is low, but 17-OHPG is only mildly, if at all, elevated.

In the very rare 17-hydroxylase deficiency, androstenedione, all other androgen-precursors (17-alpha-hydroxypregnenolone, 17-OHPG, dehydroepiandrosterone sulfate), androgens (testosterone, estrone, estradiol), and cortisol are low, while production of mineral corticoid and its precursors (particularly pregnenolone, 11-dexycorticosterone, corticosterone, and 18-hydroxycorticosterone) are increased.

For more information see Steroid Pathways.

Cautions

At birth, the hypothalamic-pituitary-adrenal axis and the hypothalamic-pituitary gonadal axis are activated, and adrenal and sex steroid levels are high. In preterm infants, the elevations can be even more pronounced due to illness and stress. As a result, preterm infants may occasionally have 17-hydroxypregnenolone levels up to 9799 ng/dL. Term infants (0-28 days) will have levels below 3104 ng/dL. These then decrease over the following 2 years to prepubertal levels (<277 ng/dL).

Clinical Reference

1. Wudy SA, Harmann M, Swoboda M. Determination of 17- hydroxypregnenolone in plasma by stabile isotope dilution/bechtop liquid chromatography-tandem mass spectrometry. Horm Res 2000;53(2):68-71
2. Therrell BL. Newborn screening for congenital adrenal hyperplasia. Endocrinol Metab Clin North Am. 2001;30(1):15-30
3. Bachega TA, Billerbeck AE, Marcondes JA, et al. Influence of different genotypes on 17-hydroxyprogesterone levels in patients with nonclassical congenital adrenal hyperplasia due to 21-hydroxylase deficiency. Clin Endocrinol. 2000;52(5):601-607
4. Kao P, Machacek DA, Magera MJ, Lacey JM, Rinaldo P. Diagnosis of adrenal cortical dysfunction by liquid chromatography-tandem mass spectrometry. Ann Clin. Lab. Sci. 2001;31:199-204
5.. Collett-Solberg PF. Congenital adrenal hyperplasia: from genetics and biochemistry to clinical practice, part I. Clin Pediatr (Phila). 2001:40(1):1-16
6. Kushnir MA, Rockwood AL, Roberts WL, Pattison EG, et al. Development and performance evaluation of a tandem mass spectrometry assay for 4 adrenal steroids. Clinical Chemistry 2006;52(8):1559-1567
7. Siklar Z, Camtosun E, Bolu S, et al. 17 alpha hydroxylase/17,20 lyase deficiency: clinical features and genetic insights from a large Turkey cohort. Endocrine. 2024;85(3):1407-1416. doi:10.1007/s12020-024-03962-6
8. Duskova M, Kolatorova L, Simkova M, et al. Steroid diagnostics of 21st century in the light of their new roles and analytical tools. Physiol Res. 2020;69(Suppl 2):S193-S203. doi:10.33549/physiolres.934517

Test Definition: 17PRN

Pregnenolone and 17-Hydroxypregnenolone, Serum

Performance

Method Description

Deuterium-labeled internal standards (pregnenolone-d4 and 17-hydroxypregnenolone-d3) are added to each sample. Pregnenolone, 17-hydroxypregnenolone, and the internal standards are extracted from the sample using solid-phase extraction. The extracts are washed, dried under nitrogen, then derivatized using hydroxylamine, and analyzed by liquid chromatography tandem mass spectrometry.(Unpublished Mayo method)

PDF Report

No

Day(s) Performed

Monday, Wednesday, Friday

Report Available

3 to 7 days

Specimen Retention Time

14 days

Performing Laboratory Location

Mayo Clinic Laboratories - Rochester Superior Drive

Fees & 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 account representative. 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. It has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information

84140-Pregnenolone
84143-17-Hydroxypregnenolone

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
17PRN	Pregnenolone and 17-OH	94303-5

	Pregnenolone	

Result ID	Test Result Name	Result LOINC® Value
81151	17-Hydroxypregnenolone, S	6765-2
88645	Pregnenolone, S	2837-3