Overview

Useful For

Confirming a clinical diagnosis of Beckwith-Wiedemann syndrome (BWS) or Russell-Silver syndrome (RSS)

Prenatal diagnosis if there is a high suspicion of BWS/RSS based on ultrasound findings or in families at risk for BWS/RSS

Genetics Test Information

This test detects deletions/duplications and determines methylation status in the *BWS*/*RSS* gene cluster.

Germline and prenatal testing are available on blood and amniocyte specimens, respectively. Prenatal testing for Beckwith-Wiedemann syndrome and Russell-Silver syndrome cannot be performed on chorionic villus specimens.

Reflex Tests

Test ID	Reporting Name	Available Separately	Always Performed
CULAF	Amniotic Fluid Culture/Genetic Test	Yes	No
MATCC	Maternal Cell Contamination, B	Yes	No
FIBR	Fibroblast Culture	Yes	No
CRYOB	Cryopreserve for Biochem Studies	No	No

Testing Algorithm

If skin biopsy is received, fibroblast culture and cryopreservation for biochemical studies will be performed at an additional charge.

For prenatal specimens only: If amniotic fluid (nonconfluent cultured cells) is received, amniotic fluid culture/genetic testÂ will be added at an additional charge. For any prenatal specimen that is received, maternal cell contamination studies will be added.

Special Instructions

- Molecular Genetics: Congenital Inherited Diseases Patient Information
- Informed Consent for Genetic Testing
- Informed Consent for Genetic Testing (Spanish)

Method Name

Methylation-Sensitive Multiplex Ligation-Dependent Probe Amplification (MLPA)

NY State Available

Yes

Specimen

Specimen Type

Varies

Additional Testing Requirements

All prenatal specimens must be accompanied by a maternal blood specimen. Order MATCC / Maternal Cell Contamination, Molecular Analysis on the maternal specimen.

Shipping Instructions

Specimen preferred to arrive within 96 hours of collection.

Prenatal specimens can be sent Monday through Thursday and **must be received by 5 p.m. Central on Friday** in order to be processed appropriately.

Specimen Required

Patient Preparation: A previous bone marrow transplant from an allogenic donor will interfere with testing. Call 800-533-1710 for instructions for testing patients who have received a bone marrow transplant.

Submit only 1 of the following specimens:

Preferred:

Specimen Type: Blood

Container/Tube:

Preferred: Lavender top (EDTA) or yellow top (ACD)

Acceptable: Any anticoagulant

Specimen Volume: 3 mL

Collection Instructions:

1. Invert several times to mix blood.

2. Send specimen in original tube.

Specimen Stability Information: Ambient (preferred)/Refrigerated/Frozen

Specimen Type: Cultured fibroblasts

Container/Tube: T-75 or T-25 flask

Specimen Volume: 1 Full T-75 or 2 full T-25 flasks

Specimen Stability Information: Ambient (preferred)/Refrigerated <24 hours

Due to the complexity of prenatal testing, consultation with the laboratory is required for all prenatal testing.

Specimen Type: Amniotic fluid

Container/Tube: Amniotic fluid container

Specimen Volume: 20 mL

Specimen Stability Information: Refrigerated (preferred)/Ambient

Acceptable:

Specimen Type: Confluent cultured cells

Container/Tube: T-25 flask

Specimen Volume: 2 flasks

Collection Instructions: Submit confluent cultured cells from another laboratory.

Specimen Stability Information: Ambient (preferred)/Refrigerated

Specimen Type: Skin biopsy

Supplies: Fibroblast Biopsy Transport Media (T115)

Container/Tube: Sterile container with any standard cell culture media (eg, minimal essential media, RPMI 1640). The solution should be supplemented with 1% penicillin and streptomycin.

Specimen Volume: 4-mm punch

Specimen Stability Information: Refrigerated (preferred)/Ambient

Forms

1. **New York Clients-Informed consent is required.** Document on the request form or electronic order that a copy is on file. The following documents are available in Special Instructions:

-Informed Consent for Genetic Testing (T576)

-Informed Consent for Genetic Testing-Spanish (T826)

2. Molecular Genetics: Congenital Inherited Diseases Patient Information(T521) in Special Instructions

Specimen Minimum Volume

Blood: 1 mL/Amniotic Fluid: 10 mL

Reject Due To

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

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Varies		

Clinical and Interpretive

Clinical Information

Beckwith-Wiedemann syndrome (BWS) is a disorder characterized by prenatal and/or postnatal overgrowth, neonatal hypoglycemia, congenital malformations, and an increased risk for embryonal tumors. Physical findings are variable and can include abdominal wall defects, macroglossia, and hemihyperplasia. The predisposition for tumor development is associated with specific tumor types such as adrenal carcinoma, nephroblastoma (Wilms tumor), hepatoblastoma, and rhabdomyosarcoma. In infancy, BWS has a mortality rate of approximately 20%.

Current data suggest that the etiology of BWS is due to dysregulation of imprinted genes in the 11p15 region of chromosome 11, including *H19* (maternally expressed), *LIT1* (official symbol *KCNQ1OT1*; paternally expressed), *IGF2* (paternally expressed), and *CDKN1C* (aliases *p57* and *KIP2*; maternally expressed). Expression of these genes is controlled by 2 imprinting centers (IC).

Approximately 85% of BWS cases appear to be sporadic, while 15% of cases are associated with an autosomal dominant inheritance pattern. When a family history is present, the etiology is often due to inherited point alterations in *CDKN1C* or an unknown cause. The etiology of sporadic cases includes:

-Hypomethylation of imprinting center 2 (IC2) (*LIT1)*: approximately 50% to 60%

-Paternal uniparental disomy of chromosome 11: approximately 10% to 20%

-Hypermethylation of imprinting center 1 (IC1) (*H19)*: approximately 2% to 7%

-Unknown: approximately 10% to 20%

-Point alteration in *CDKN1C*: approximately 5% to 10%

-Cytogenetic abnormality: approximately 1% to 2%

-Differentially methylated region 1 (DMR1) or DMR2 microdeletion: rare

The clinical presentation of BWS is dependent on which gene in the 11p15 region is involved. The risk for cancer has been shown to be significantly higher in patients with abnormal methylation of IC1 (*H19)* versus IC2 (*LIT1)*. In patients with abnormal methylation of IC2 (*LIT1)*, abdominal wall defects and overgrowth are seen at a higher frequency.

Russell-Silver syndrome (RSS) is a rare genetic condition with an incidence of approximately 1 in 100,000. RSS is characterized by pre- and postnatal growth retardation with normal head circumference, characteristic facies, fifth finger clinodactyly, and asymmetry of the face, body, and/or limbs. Less commonly observed clinical features include cafe au lait spots, genitourinary anomalies, motor, speech, cognitive delays, and hypoglycemia. Although clinical diagnostic criteria have been developed, it has been demonstrated that many patients with molecularly confirmed RSS do not meet strict clinical diagnostic criteria for RSS. Therefore, most groups recommend a relatively low threshold for considering molecular testing in suspected cases of RSS.

RSS is a genetically heterogeneous condition that is associated with genetic and epigenetic alterations at chromosome 7 and the chromosome 11p15.5 region. The majority of cases of RSS are sporadic, although familial cases have been reported. The etiology of sporadic cases of RSS includes:

-Hypomethylation of IC1 (*H19*): approximately 30% to 50%

-Maternal uniparental disomy (UPD) of chromosome 7: approximately 5% to 10%*

-11p15.5 duplications: rare

-Chromosome 7 duplications: rare*

*Note that this test does not detect chromosome 7 UPD. However, testing is available; order UNIPD / Uniparental Disomy, Varies.

The clinical phenotype of RSS has been associated with the specific underlying molecular etiology. Patients with hypomethylation of IC1 (*H19*) are more likely to exhibit "classic" RSS phenotype (ie, severe intrauterine growth retardation, postnatal growth retardation, and asymmetry), while patients with maternal UPD7 often show a milder clinical phenotype. Despite these general genotype-phenotype correlations, many exceptions have been reported.

Methylation abnormalities of IC1 (*H19*) and IC2 (*LIT1*) can be detected by methylation-sensitive multiple ligation-dependent probe amplification. While testing can determine methylation status, it does not identify the mechanism responsible for the methylation defect (such as paternal uniparental disomy or cytogenetic abnormalities). Hypomethylation of IC2 (*LIT1)* is hypothesized to silence the expression of a number of maternally expressed genes, including *CDKN1C*. Hypermethylation of IC1 is hypothesized to silence the expression of *H19*, while also resulting in overexpression of *IGF2*. Absence of *CDKN1C* and *H19* expression, in addition to overexpression of *IGF2*, is postulated to contribute to the clinical phenotype of BWS. Hypomethylation of IC1 is hypothesized to result in overexpression of *H19* and underexpression of the *IGF2*, which is thought to contribute to the clinical phenotype of RSS.

Reference Values

An interpretive report will be provided.

Interpretation

An interpretive report will be provided.

Cautions

In addition to disease-related probes, the multiple ligation-dependent probe amplification technique utilizes probes localized to other chromosomal regions as internal controls. In certain circumstances, these control probes may detect other diseases or conditions for which this test was not specifically intended. Results of the control probes are not normally reported. However, in cases where clinically relevant information is identified, the ordering physician will be informed of the result and provided with recommendations for any appropriate follow-up testing.

Rare alterations exist that could lead to false-negative or false-positive results. If results obtained do not match the clinical findings, additional testing should be considered.

Test results should be interpreted in the context of clinical findings, family history, and other laboratory data. Errors in the interpretation of results may occur if information given is inaccurate or incomplete.

Methylation status cannot be assessed on chorionic villus specimens.

This assay does not detect maternal uniparental disomy of chromosome 7 or cytogenetic abnormalities such as translocations, inversions, or duplications.

Supportive Data

Normal methylation index was derived by studying 150 normal individuals. For 65 patients referred for Beckwith-Wiedemann syndrome testing, results of this multiple ligation-dependent probe amplification (MLPA) assay were compared to a Southern blot method. Results were concordant for 64 of 65 specimens. In 1 specimen, a deletion was identified by MLPA that was not detected by the Southern blot method. For 55 patients referred for Russell-Silver syndrome testing, results of this MLPA assay were compared to *H19* Southern blot. Results were concordant for 53 of 55 specimens. Two specimens, both amniotic fluid, were positive for a *H19* hypomethylation defect by Southern blot that was not detected by MLPA.

Clinical Reference

1. DeBaun MR, Niemitz EL, McNeil DE, Brandenburg SA, Lee MP, Feinberg AP: Epigenetic alterations of *H19* and *LIT1* distinguish patients with Beckwith-Wiedemann Syndrome with cancer and birth defects. Am J Hum Genet. 2002;70:604-611

2. Choufani S, Shuman C, Weksberg R: Beckwith-Wiedemann Syndrome. Am J Med Genet C Semin Med Genet. 2010;154C:343-354

3. Wakeling EL: Silver-Russell syndrome. Arch Dis Child. 2011;96(12):1156-1161

4. Eggermann T, Begemann M, Binder G, Spengler S: Silver-Russell syndrome: genetic basis and molecular genetic testing. Orphanet J Rare Dis. 2010;5:19-26

5. Priolo M, Sparago A, Mammi C, Cerrato F, Lagana C, Riccio A: MS-MLPA is a specific and sensitive technique for detecting all chromosome 11p15.5 imprinting defects of BWS and SRS in a single-tube experiment. Eur J Hum Genet. 2008;16:565-571

Performance

Method Description

Methylation-sensitive multiple ligation-dependent probe amplification is utilized to test for the presence of large deletions, duplications, and methylation defects in the imprinting center 1 (IC1) (*H19*) and IC2 (*LIT1*) critical regions on chromosome 11p15.(Unpublished Mayo method)

PDF Report

No

Day(s) and Time(s) Test Performed

Monday; 10 a.m.

Specimens received Monday through Thursday will be run on the following Monday. Specimens received Friday through Sunday will be set up a week from the following Monday.

Analytic Time

14 days

Maximum Laboratory Time

21 days

Specimen Retention Time

Whole Blood: 2 weeks (if available); Extracted DNA: 3 months

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

81401-H19 (imprinted maternally expressed transcript [non-protein coding]) (eg, Beckwith-Wiedemann syndrome), methylation analysis

81401-KCNQ1OT1 (KCNQ1 overlapping transcript 1 [non-protein coding]) (eg, Beckwith-Wiedemann syndrome) methylation analysisÂ

88233-Tissue culture, skin or solid tissue biopsy (if appropriate)

88240-Cryopreservation (if appropriate)

88235-Tissue culture for amniotic fluid (if appropriate)

81265-Comparative analysis using Short Tandem Repeat (STR) markers; patient and comparative specimen (eg, pre-transplant recipient and donor germline testing, post-transplant non-hematopoietic recipient germline [eg, buccal swab or other germline tissue sample] and donor testing, twin zygosity testing or maternal cell contamination of fetal cells (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC Value
BWRS	BWS/RSS Molecular Analysis	In Process

Result ID	Test Result Name	Result LOINC Value
52845	Result Summary	50397-9
52846	Result	82939-0
52847	Interpretation	69047-9
52848	Reason for Referral	42349-1
52849	Specimen	31208-2
52850	Source	31208-2
52851	Released By	18771-6