Overview

Useful For
Providing a comprehensive genetic evaluation for patients with a personal or family history suggestive of telomeropathies

Establishing a diagnosis of a telomeropathy, in some cases, allowing for appropriate management and surveillance for disease features

Identifying pathogenic variants within genes known to be associated with increased risk for telomere defects allowing for predictive testing of at-risk family members

Genetics Test Information
This test includes next-generation sequencing and supplemental Sanger sequencing to evaluate for the genes listed on the panel.

Highlights
This test uses next-generation sequencing to test for variants in the CTC1, DKC1, NHP2, NOP10, RTEL1, TERC, TERT, TINF2, USB1 (C16ORF57), and WRAP53 (TCAB1) genes.

Identification of a pathogenic variant may assist with prognosis, clinical management, familial screening, and genetic counseling.

Reflex Tests

<table>
<thead>
<tr>
<th>Test ID</th>
<th>Reporting Name</th>
<th>Available Separately</th>
<th>Always Performed</th>
</tr>
</thead>
<tbody>
<tr>
<td>FIBR</td>
<td>Fibroblast Culture</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>CRYOB</td>
<td>Cryopreserve for Biochem Studies</td>
<td>No</td>
<td>No</td>
</tr>
</tbody>
</table>

Testing Algorithm
For skin biopsy or cultured fibroblast specimens, fibroblast culture and cryopreservation testing will be performed at an additional charge. If viable cells are not obtained, the client will be notified.

Special Instructions
- Informed Consent for Genetic Testing
- Primary Immunodeficiencies Patient Information

Method Name
Custom Sequence Capture and Targeted Next-Generation Sequencing followed by Polymerase Chain Reaction (PCR) and Supplemental Sanger Sequencing

NY State Available
Yes

Specimen
Specimen Type
Varies

Ordering Guidance
This test in not for measuring telomere length. This test is a second-tier genetic test for hereditary telomere defects.

Targeted testing for familial variants (also called site-specific or known mutations testing) is available for the genes on this panel. See FMTT / Familial Mutation, Targeted Testing, Varies.

Necessary Information

1. **Primary Immunodeficiencies Patient Information** (T791) is strongly recommended, but not required, to be filled out and sent with the specimen. This information aids in providing a more thorough interpretation of test results. Ordering providers are strongly encouraged to complete the form and send it with the specimen.

2. Include physician name and phone number with specimen.

Specimen Required

**Patient Preparation:** A previous bone marrow transplant from an allogenic donor or a recent (ie, <6 weeks from time of sample collection) heterologous blood transfusion 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:** Whole blood

**Container/Tube:** Lavender top (EDTA)

**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

**Specimen Type:** Blood spot

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

**Container/Tube:**

**Preferred:** Collection card (Whatman Protein Saver 903 Paper)

**Acceptable:** Whatman FTA Classic paper, Ahlstrom 226 filter paper, or Blood Spot Collection Card (T493)
**Specimen Volume:** 2 to 5 blood spots on collection card

**Collection Instructions:**

1. An alternative blood collection option for a patient <1 year of age is finger stick.
2. Let blood dry on the filter paper at ambient temperature in a horizontal position for 3 hours.
3. Do not expose specimen to heat or direct sunlight.
4. Do not stack wet specimens.
5. Keep specimen dry.

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

**Specimen Type:** Peripheral blood mononuclear cells (PBMCs)

**Container/Tube:** Cell pellet

**Collection Instructions:** Send as a suspension in freezing medium or cell pellet frozen on dry ice.

**Specimen Stability Information:** 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

**Additional Information:** Indicate the tests to be performed on the fibroblast culture cells. A separate culture charge will be assessed under FIBR / Fibroblast Culture. An additional 4 weeks is required to culture fibroblasts before genetic testing can occur.

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

**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. Tubes of culture media can be supplied upon request (Eagle's minimum essential medium with 1% penicillin and streptomycin [T115]).

**Specimen Volume:** 4-mm punch

**Additional Information:** A separate culture charge will be assessed under FIBR / Fibroblast Culture. An additional 4 weeks is required to culture fibroblasts before genetic testing can occur.
**Specimen Stability Information:** Refrigerated (preferred)/Ambient

**Specimen Type:** DNA

**Container/Tube:** 2 mL screw top tube

**Specimen Volume:** 100 mcL (microliters)

**Collection Instructions:**
1. The preferred volume is 100 mcL at a concentration of 250 ng/mcL.
2. Include concentration and volume on tube.

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

**Forms**
1. **New York Clients-Informed consent is required.** Please document on the request form or electronic order that a copy is on file. An Informed Consent for Genetic Testing (T576) is available in Special Instructions.

2. **Primary Immunodeficiencies Patient Information (T791) is required.** See Special Instructions.

**Specimen Minimum Volume**
Whole blood: 1 mL

**Reject Due To**

**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>Varies</td>
<td>Varies</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Clinical and Interpretive**

**Clinical Information**

Telomeres are highly specialized structures composed of TTAGGG nucleotide repeats and proteins that protect chromosome ends. Under normal circumstances, telomeres shorten with every cycle of DNA replication. Telomerase is an enzyme complex that can extend the length of the telomere, thus helping to slow the shortening process. Telomerase is most active in highly proliferative tissues such as lymphocytes, skin, intestine, and bone marrow. Variants in genes involved with telomere repair and maintenance may cause telomeres to shorten more quickly than normal.

Telomere biology disorders (TBD) are a complex group of bone marrow failure syndromes characterized by abnormally short telomeres. The severity of these syndromes is variable, and they may present in children or adults. In addition to bone marrow failure, other symptoms of telomere biology disorders include pulmonary fibrosis, liver disease, gastrointestinal disease, and mucocutaneous abnormalities. Recognition and diagnosis of underlying TBD
is important as it can help guide treatment decisions.

Dyskeratosis congenita (DC) was the first TBD to be described. The subsets of DC include classic DC, Hoyeraal Hreidarsson syndrome (HHS), Revesz syndrome, DC-like conditions, and isolated subtypes. Patients with the classic forms of DC are usually diagnosed in childhood, and they have a triad of mucocutaneous features including dysplastic nails, anomalies of skin pigmentation, and oral leukoplakia. Other features of DC may include bone marrow failure, gastrointestinal disease, liver disease, pulmonary fibrosis, a predisposition to certain cancers, and other medical problems. Other TBD presenting in childhood include HHS and Revesz syndrome.

TBD may also manifest in adulthood and the presentation can be variable. A broad umbrella of conditions could include bone marrow failure, pulmonary fibrosis, liver disease not otherwise classified, myelodysplastic syndrome, acute myeloid leukemia, or early onset of malignancies within the DC grouping.

Telomere biology disorders can be inherited in a variety of patterns, including X-linked recessive, autosomal dominant, and autosomal recessive. At least 50% of patients with DC have mutations in the DCK1, TERC, TERT, TINF2, NHP2, and NOP10 genes. In autosomal dominant DC, phenotypes may present at a younger age and more severely in successive generations (genetic anticipation). See Table for a summary of the genes included in this panel, associated diseases, and the mode of inheritance.

Alternatively, some patients may have 1 of these 3 features along with a hypocellular bone marrow. These patients all have very short telomeres (<1% percentile of age) in leukocytes. Patients with HHS have the features of classic DC but additionally have cerebellar hypoplasia. Telomere length analysis in leukocyte subsets is performed by flow-FISH (see references). They can also have low T cell numbers with severe B and NK cell lymphopenia (T+/−B-NK−) reminiscent of severe combined immunodeficiency (SCID). In Revesz syndrome, patients have bilateral exudative retinopathy along with other features of DC.

In patients who do not meet the diagnostic criteria of DC but have several features reminiscent of the disease, a classification of DC-like may be applied. This could include presence of bone marrow failure, developmental delay, familial history of pulmonary fibrosis, and no other clear diagnosis. These patients usually have short, but not very short telomeres, and may or may not have a genetic defect in one of the known telomere biology genes. However, these patients need careful monitoring as they may evolve into the more classic forms of DC over time. Individuals who have mutations in one of the telomere genes and have very short telomeres (<1% percentile of normal for age) who have a single feature of a telomere disorder or DC could be considered to be an isolated subtype. The stringency of monitoring depends on the individual case, age of patient, complications, and should include counseling for family members for potential disease risk and the phenomenon of genetic anticipation.

Patients with aplastic anemia may have mutations in TERC, TERT, and TINF2 genes. Approximately 20% of patients with idiopathic pulmonary fibrosis (IPF) have a familial inheritance, which is autosomal dominant but with variable penetrance and approximately 30% of patients with familial IPF have short telomeres with genetic defects in the telomere genes. The genes most commonly identified in the context of IPF are TERC and TERT.

The telomerase complex includes a reverse transcriptase encoded by TERT, RNA template (encoded by TERC), and associated other proteins that regulate the assembly, trafficking, recruitment of telomerase to telomeres and stability of telomeres, including dyskerin (DKC1). Other members of the telomerase complex include NOP10 (NOLA3) and NHP2 (NOLA2). The shelterin complex, which is a 6-protein complex that coats telomeres and offers telomere end protection. The shelterin complex directs telomere length homeostasis (T-loop) and prevents DNA damage response activation. The DNA helicase, RTEL1 promotes telomere elongation through the unwinding of the T-loop. TCAF1 (WRAP53) directs trafficking of the telomerase complex to the telomeric ends. The CST complex of 3 proteins (CTC1 and others) inhibits telomerase activity and promotes capping.

Table. Genes included in this panel
<table>
<thead>
<tr>
<th>Gene (alias)</th>
<th>Protein</th>
<th>OMIM</th>
<th>Incidence</th>
<th>Inheritance</th>
<th>Phenotype disorder</th>
</tr>
</thead>
<tbody>
<tr>
<td>CTC1</td>
<td>CST complex subunit CTC1</td>
<td>613129</td>
<td>Approximately 1-3% of DC</td>
<td>AR</td>
<td>Cerebroretinal microangiopathy with calcifications and cysts</td>
</tr>
<tr>
<td>DKC1</td>
<td>H/ACA ribonucleoprotein complex subunit 4 isoform 1</td>
<td>300126</td>
<td>Approximately 17-36% of DC</td>
<td>XL</td>
<td>Dyskeratosis congenita</td>
</tr>
<tr>
<td>NHP2</td>
<td>H/ACA ribonucleoprotein complex subunit 2 isoform a</td>
<td>606470</td>
<td>&lt;1% of DC</td>
<td>AR</td>
<td>Dyskeratosis congenita</td>
</tr>
<tr>
<td>NOP10</td>
<td>H/ACA ribonucleoprotein complex subunit 3</td>
<td>606471</td>
<td>&lt;1% of DC</td>
<td>AR</td>
<td>Dyskeratosis congenita</td>
</tr>
<tr>
<td>RTEL1</td>
<td>Regulator of telomere elongation helicase 1 isoform 2</td>
<td>608833</td>
<td>Rare</td>
<td>AR, AD</td>
<td>Dyskeratosis congenita, pulmonary fibrosis and/or bone marrow failure, dyskeratosis congenita</td>
</tr>
<tr>
<td>TERC</td>
<td>Telomerase RNA component</td>
<td>602322</td>
<td>Approximately 6-10% of DC</td>
<td>AD</td>
<td>Dyskeratosis congenita, aplastic anemia, susceptibility to idiopathic pulmonary fibrosis</td>
</tr>
<tr>
<td>TERT</td>
<td>Telomerase reverse transcriptase isoform 1</td>
<td>187270</td>
<td>Approximately 1-7% of DC</td>
<td>AR,AD</td>
<td>Dyskeratosis congenita, acute myeloid leukemia, cutaneous malignant melanoma, pulmonary fibrosis and/or bone marrow failure, telomere-related</td>
</tr>
<tr>
<td>TINF2</td>
<td>TERF1-interacting nuclear factor 2 isoform 1</td>
<td>604319</td>
<td>Approximately 11-24% of DC</td>
<td>AD</td>
<td>Dyskeratosis congenita, Revesz syndrome</td>
</tr>
<tr>
<td>USB1</td>
<td>U6 snRNA phosphodiesterase isoform 1</td>
<td>613276</td>
<td>Rare</td>
<td>AR</td>
<td>Poikiloderma with neutropenia, A-A</td>
</tr>
<tr>
<td>WRAP53</td>
<td>Telomerase Cajal body protein 1 (TCAB1)</td>
<td>612661</td>
<td>Approximately 3% of DC</td>
<td>AR</td>
<td>Dyskeratosis congenita</td>
</tr>
</tbody>
</table>
**Reference Values**
An interpretive report will be provided.

**Interpretation**
Evaluation and categorization of variants is performed using the most recent published American College of Medical Genetics and Genomics (ACMG) recommendations as a guideline. Variants are classified based on known, predicted, or possible pathogenicity and reported with interpretive comments detailing their potential or known significance.

Multiple in silico evaluation tools may be used to assist in the interpretation of these results. The accuracy of predictions made by in silico evaluation tools is highly dependent upon the data available for a given gene, and predictions made by these tools may change over time. Results from in silico evaluation tools should be interpreted with caution and professional clinical judgment.

Unless reported or predicted to cause disease, alterations found deep in the intron or alterations that do not result in an amino acid substitution are not reported. Information about these variants and common polymorphisms are available upon request.

The telomerase database is a useful tool for variant review and classification of telomere disorders that may be used in some cases.

**Cautions**

**Clinical Correlations:**
Some individuals who have involvement of one or more of the genes on the panel may have a variant that is not identified by the methods performed (eg, promoter variants, deep intronic variants). The absence of a variant, therefore, does not eliminate the possibility of disease. Test results should be interpreted in context of clinical findings, family history, and other laboratory data. Misinterpretation of results may occur if the information provided is inaccurate or incomplete.

Â

For predictive testing of asymptomatic individuals, it is important to first document the presence of a gene variant in an affected family member.

Â

Short telomeres may be present in leukocytes or buccal cells with a variety of malignancies. Short telomeres have also been reported in nonmalignant disorders such as metabolic conditions, cardiovascular disease, diabetes, and in smokers. At least half of patients with a BMFS or DC phenotype do not have a detectable variant in the 10 genes tested in the panel and, therefore, there may be additional genes that need to be discovered. Most cases of telomere disorders appear to occur sporadically with no prior family history, and this could be related to incomplete penetrance, variable expressivity, or de novo variants. It is important to correlate the genetic testing data with clinical phenotype and other relevant testing, including assessment of telomere length.

Â

**Technical Limitations:**
Next-generation sequencing may not detect all types of genetic variants. Additionally, rare alterations (ie,
polymorphisms) may be present that could lead to false-negative or false-positive results. The variant detection software has lower detection efficiency for insertion/deletion variants as compared to single nucleotide variants. Therefore, small deletions and insertions greater than 8 nucleotides in length may not be detected by this test. Copy number variations (CNV) are not currently reported for any of the genes on this panel. Additionally, rare polymorphisms may be present that could lead to false-negative or false-positive results. In some cases, DNA variants of undetermined significance may be identified. If a diagnosis of one of the syndromes on this panel is still suspected, consider a Laboratory Director or genetic counselor consultation. 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.

Reclassification of Variants-Policy:

At this time, it is not standard practice for the laboratory to systematically review likely pathogenic variants or variants of uncertain significance that are detected and reported. The laboratory encourages health care providers to contact the laboratory at any time to learn how the status of a particular variant may have changed over time. Consultation with a healthcare provider, or team of healthcare providers, with expertise in genetics and primary immunodeficiencies, is recommended for interpretation of this result.

A list of benign and likely benign variants detected for this patient is available from the lab upon request.

Contact the laboratory if additional information is required regarding the transcript or human genome assembly used for the analysis of this patient's results.

Clinical Reference

1. Podlevsky JD: The Telomere Database. NAR 2008;36:D339-D343

Performance
Method Description

Next-generation sequencing (NGS) is performed using an Illumina instrument with paired-end reads. The DNA is prepared for NGS using a custom Agilent Sure Select Target Enrichment System. Data is analyzed with a bioinformatics software pipeline. Supplemental Sanger sequencing may be performed occasionally in regions where NGS is insufficient for data capture or not specific enough to correctly identify a variant. (Unpublished Mayo method)

Genes analyzed: CTC1, DKC1, NHP2, NOP10, RTE1, TERC, TERT, TINF2, USB1(C16ORF57), WRAP53 (TCAB1)

PDF Report
No

Day(s) Performed
Monday

Report Available
6 to 8 weeks

Specimen Retention Time
Extracted DNA: 2 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 US Food and Drug Administration.

CPT Code Information
81479

LOINC® Information

<table>
<thead>
<tr>
<th>Test ID</th>
<th>Test Order Name</th>
<th>Order LOINC Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>TELGP</td>
<td>Telomere Defects Gene Panel</td>
<td>In Process</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>BA3918</td>
<td>Gene(s) Evaluated</td>
<td>48018-6</td>
</tr>
<tr>
<td>BA3919</td>
<td>Result Summary</td>
<td>50397-9</td>
</tr>
<tr>
<td>BA3920</td>
<td>Result Details</td>
<td>82939-0</td>
</tr>
<tr>
<td>Result ID</td>
<td>Test Result Name</td>
<td>Result LOINC Value</td>
</tr>
<tr>
<td>-----------</td>
<td>----------------------------</td>
<td>--------------------</td>
</tr>
<tr>
<td>BA3921</td>
<td>Interpretation</td>
<td>69047-9</td>
</tr>
<tr>
<td>BA3922</td>
<td>Additional Information</td>
<td>48767-8</td>
</tr>
<tr>
<td>BA3923</td>
<td>Method</td>
<td>85069-3</td>
</tr>
<tr>
<td>BA3924</td>
<td>Disclaimer</td>
<td>62364-5</td>
</tr>
<tr>
<td>BA3925</td>
<td>Reviewed by</td>
<td>18771-6</td>
</tr>
</tbody>
</table>