MAYO CLINIC LABORATORIES

Test Definition: DMITO

Mitochondrial DNA Deletion Heteroplasmy, ddPCR, Varies

Overview

Useful For

Assessing the heteroplasmy level of previously detected large mitochondrial DNA (mtDNA) deletions.

Screening family members for previously detected large mtDNA deletions.

This test **is not recommended for** first tier diagnostic testing for mitochondrial disorders.

This test **does not assess** mtDNA depletion.

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
_STR1	Comp Analysis using STR (Bill only)	No, (Bill only)	No
_STR2	Add'l comp analysis w/STR (Bill Only)	No, (Bill only)	No
CULFB	Fibroblast Culture for Genetic Test	No	No
CULAF	Amniotic Fluid Culture/Genetic Test	Yes	No
MATCC	Maternal Cell Contamination, B	Yes	No

Genetics Test Information

This test utilizes droplet digital polymerase chain reaction (ddPCR) for confirmation and determination of heteroplasmy levels of previously detected large mitochondrial DNA (mtDNA) deletions.

Identification of heteroplasmy for large mtDNA deletions may assist with diagnosis, prognosis, clinical management, recurrence risk assessment, familial screening, and genetic counseling for mtDNA deletion syndromes.

Testing Algorithm

Skin biopsy:
If skin biopsy is received, fibroblast culture will be added at an additional charge. If viable cells are not obtained, the client will be notified.

Prenatal specimens:
If an amniotic fluid specimen is received, an amniotic fluid culture will be performed at an additional charge.
If chorionic villi, cultured chorionic villi, or cultured amniocyte specimen is received, a fibroblast culture will be performed at an additional charge.
For any prenatal specimen that is received, maternal cell contamination testing will be performed at an additional

charge.

Cord blood:
For cord blood specimens that have an accompanying maternal blood specimen, maternal cell contamination studies will be performed at an additional charge.

Special Instructions
- Muscle Biopsy Specimen Preparation Instructions
- Molecular Genetics: Biochemical Disorders Patient Information
- Informed Consent for Genetic Testing
- Blood Spot Collection Card-Spanish Instructions
- Blood Spot Collection Card-Chinese Instructions
- Informed Consent for Genetic Testing (Spanish)
- Blood Spot Collection Instructions

Method Name
Droplet Digital Polymerase Chain Reaction (ddPCR)

NY State Available
Yes

Specimen

Specimen Type
Varies

Ordering Guidance
For diagnosis of a mitochondrial DNA deletion syndrome, the recommended first tier test is MITOP/ Mitochondrial Full Genome Analysis, Next-Generation Sequencing (NGS), Varies.

Specimen Required
Patient Preparation: A previous hematopoietic stem cell transplant from an allogenic donor will interfere with testing. For information about testing patients who have received a hematopoietic stem cell transplant, call 800-533-1710.

Submit only 1 of the following specimens:

Specimen Type: Whole blood
Container/Tube:
Preferred: Lavender top (EDTA) or yellow top (ACD)
Acceptable: Green top (Sodium heparin)
Specimen Volume: 3 mL
Collection Instructions:
1. Invert several times to mix blood.

2. Send whole blood specimen in original tube. **Do not aliquot**.
3. Whole blood collected postnatal from an umbilical cord is also acceptable. See Additional Information
Specimen Stability Information: Ambient 4 days/Refrigerated 4 days/Frozen 4 days
Additional Information:
1. Specimens are preferred to be received within 4 days of collection. Extraction will be attempted for specimens received after 4 days, and DNA yield will be evaluated to determine if testing may proceed.
2. To ensure minimum volume and concentration of DNA are met, the requested volume must be submitted. Testing may be canceled if DNA requirements are inadequate.
3. For postnatal umbilical cord whole blood specimens, maternal cell contamination studies are recommended to ensure test results reflect that of the patient tested. A maternal blood specimen is required to complete maternal cell contamination studies. Order MATCC / Maternal Cell Contamination, Molecular Analysis, Varies on both the cord blood and maternal blood specimens under separate order numbers.

Specimen Type: Saliva
Patient Preparation: Patient should not eat, drink, smoke, or chew gum 30 minutes prior to collection.
Supplies:
DNA Saliva Kit High Yield (T1007)
Saliva Swab Collection Kit (T786)
Container/Tube:
Preferred: High-yield DNA saliva kit
Acceptable: Saliva swab
Specimen Volume: 1 Tube if using T1007 or 2 swabs if using T786
Collection Instructions: Collect and send specimen per kit instructions.
Specimen Stability Information: Ambient (preferred) 30 days/Refrigerated 30 days
Additional Information: Saliva specimens are acceptable but not recommended. Due to lower quantity/quality of DNA yielded from saliva, some aspects of the test may not perform as well as DNA extracted from a whole blood sample. When applicable, specific gene regions that were unable to be interrogated will be noted in the report. Alternatively, additional specimen may be required to complete testing.

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: Ambient (preferred) <24 hours/Refrigerated <24 hours
Additional Information:
1. Specimens are preferred to be received within 24 hours of collection. Culture and extraction will be attempted for specimens received after 24 hours and will be evaluated to determine if testing may proceed.
2. A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing. An additional 3 to 4 weeks are required to culture fibroblasts before genetic testing can occur.

Specimen Type: Cultured fibroblasts
Source: Skin
Container/Tube: T-25 flask

Specimen Volume: 2 Flasks
Collection Instructions: Submit confluent cultured fibroblast cells from a tissue biopsy.
Specimen Stability Information: Ambient (preferred) <24 hours/Refrigerated <24 hours
Additional Information:
1. Specimens are preferred to be received within 24 hours of collection. Culture and extraction will be attempted for specimens received after 24 hours and will be evaluated to determine if testing may proceed.
2. A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing. An additional 3 to 4 weeks are required to culture fibroblasts before genetic testing can occur.

Specimen Type: Tissue biopsy
Supplies: Hank's Solution (T132)
Container/Tube: Sterile container with sterile Hank's balanced salt solution, Ringer's solution, or normal saline
Specimen Volume: 0.5 to 3 cm(3) or larger
Specimen Stability Information: Ambient (preferred) <24 hours/Refrigerated <24 hours
Additional Information:
1. Specimens are preferred to be received within 24 hours of collection. Culture and extraction will be attempted for specimens received after 24 hours and will be evaluated to determine if testing may proceed.
2. A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing. An additional 3 to 4 weeks are required to culture fibroblasts before genetic testing can occur.

Specimen Type: Muscle tissue biopsy
Supplies: Muscle Biopsy Kit (T541)
Specimen Volume: 20 to 80 mg
Collection Instructions: Prepare and transport specimen per instructions in Muscle Biopsy Specimen Preparation.
Specimen Stability Information: Frozen (preferred) <24 hours/Ambient <24 hours/Refrigerated <24 hours
Additional Information: Specimens are preferred to be received within 24 hours of collection. Extraction will be attempted for specimens received after 24 hours and will be evaluated to determine if testing may proceed.

Specimen Type: Snap frozen nerve tissue biopsy
Collection Instructions: Prepare snap frozen tissue biopsy per surgical procedure
Specimen Volume: 0.25 to 0.5 cm
Specimen Stability Information: Frozen

Specimen Type: Blood spot
Supplies: Card-Blood Spot Collection (Filter Paper) (T493)
Container/Tube:
Preferred: Collection card (Whatman Protein Saver 903 Paper)
Acceptable: PerkinElmer 226 filter paper or blood spot collection card
Specimen Volume: 2 to 5 Blood spots
Collection Instructions:
1. An alternative blood collection option for a patient older than 1 year is a fingerstick. For detailed instructions, see How to Collect a Dried Blood Spot Sample.
2. Let blood dry on the filter paper at ambient temperature in a horizontal position for a minimum of 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

Additional Information:

1. Blood spot specimens are acceptable but not recommended. Multiple extractions will be required to obtain sufficient yield for supplemental analysis, and there is significant risk for test failure due to insufficient DNA.
2. Due to lower concentration of DNA yielded from blood spot, some aspects of the test may not perform as well as DNA extracted from a whole blood sample. When applicable, specific gene regions that were unable to be interrogated will be noted in the report. Alternatively, additional specimen may be required to complete testing.
3. For collection instructions, see Blood Spot Collection Instructions
4. For collection instructions in Spanish, see Blood Spot Collection Card-Spanish Instructions (T777)
5. For collection instructions in Chinese, see Blood Spot Collection Card-Chinese Instructions (T800)

Specimen Type: Extracted DNA

Container/Tube:

Preferred: Screw Cap Micro Tube, 2 mL with skirted conical base

Acceptable: Matrix tube, 1 mL

Collection Instructions:

1. The preferred volume is at least 100 mcL at a concentration of 75 ng/mcL.
2. Include concentration and volume on tube.

Specimen Stability Information: Frozen (preferred) 1 year/Ambient/Refrigerated

Additional Information: DNA must be extracted in a CLIA-certified laboratory or equivalent and must be extracted from a specimen type listed as acceptable for this test (including applicable anticoagulants). Our laboratory has experience with Chemagic, Puregene, Autopure, MagnaPure, and EZ1 extraction platforms and cannot guarantee that all extraction methods are compatible with this test. If testing fails, one repeat will be attempted, and if unsuccessful, the test will be reported as failed and a charge will be applied. If applicable, specific gene regions that were unable to be interrogated due to DNA quality will be noted in the report.

PRENATAL SPECIMENS

Due to its complexity, consultation with the laboratory is required for all prenatal testing; call 800-533-1710 to speak to a genetic counselor.

Specimen Type: Amniotic fluid

Container/Tube: Amniotic fluid container

Specimen Volume: 20 mL

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

Additional Information: Specimen will only be tested after culture.

1. Specimens are preferred to be received within 24 hours of collection. Culture and extraction will be attempted for specimens received after 24 hours and will be evaluated to determine if testing may proceed.
2. A separate culture charge will be assessed under CULAF / Culture for Genetic Testing, Amniotic Fluid. An additional 2 to 3 weeks are required to culture amniotic fluid before genetic testing can occur.
3. **All prenatal specimens must be accompanied by a maternal blood specimen;** order MATCC / Maternal Cell Contamination, Molecular Analysis, Varies on the maternal specimen.

Specimen Type: Confluent cultured amniocytes
Container/Tube: T-25 flask
Specimen Volume: 2 Flasks
Collection Instructions: Submit confluent cultured amniocytes from another laboratory
Specimen Stability Information: Ambient (preferred) <24 hours/Refrigerated <24 hours
Additional Information:
1. Specimens are preferred to be received within 24 hours of collection. Culture and extraction will be attempted for specimens received after 24 hours and will be evaluated to determine if testing may proceed.
2. A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing.
3. **All prenatal specimens must be accompanied by a maternal blood specimen;** order MATCC / Maternal Cell Contamination, Molecular Analysis, Varies on the maternal specimen.

Specimen Type: Chorionic villi
Container/Tube: 15-mL tube containing 15 mL of transport media
Specimen Volume: 20 mg
Specimen Stability Information: Ambient (preferred) <24 hours/Refrigerated <24 hours
Additional Information: Specimen will only be tested after culture.
1. Specimens are preferred to be received within 24 hours of collection. Culture and extraction will be attempted for specimens received after 24 hours and will be evaluated to determine if testing may proceed.
2. A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing. An additional 3 to 4 weeks are required to culture fibroblasts before genetic testing can occur.
3. **All prenatal specimens must be accompanied by a maternal blood specimen;** order MATCC / Maternal Cell Contamination, Molecular Analysis, Varies on the maternal specimen.

Specimen Type: Cultured chorionic villi
Container/Tube: T-25 flasks
Specimen Volume: 2 Full flasks
Collection Instructions: Submit confluent cultured cells from another laboratory.
Specimen Stability Information: Ambient (preferred) <24 hours/Refrigerated <24 hours
Additional Information:
1. Specimens are preferred to be received within 24 hours of collection. Culture and extraction will be attempted for specimens received after 24 hours and will be evaluated to determine if testing may proceed.
2. A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing.
3. **All prenatal specimens must be accompanied by a maternal blood specimen;** order MATCC / Maternal Cell Contamination, Molecular Analysis, Varies on the maternal specimen.

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:
-Informed Consent for Genetic Testing (T576)
-Informed Consent for Genetic Testing (Spanish) (T826)
2. Molecular Genetics: Biochemical Disorders Patient Information (T527)
3. If not ordering electronically, complete, print, and send 1 of the following forms with the specimen:
-Neurology Specialty Testing Client Test Request (T732)

-Biochemical Genetics Test Request (T798)

Specimen Minimum Volume

See Specimen Required

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 & Interpretive

Clinical Information

Large deletions in the mitochondrial genome (mtDNA deletions) cause up to 10% of primary mitochondrial disease.(1) mtDNA deletions typically present with 1 of 3 syndromes, but a large amount of clinical overlap exists. The 3 syndromes include Kearns-Sayre syndrome, Pearson syndrome, and progressive external ophthalmoplegia (PEO). Occasionally large mtDNA deletions may cause Leigh syndrome. The phenotypes for these conditions vary.

Kearns-Sayre syndrome typically has an age of onset of less than 20 years and is characterized by pigmentary retinopathy or PEO, cardiac conduction defects, ataxia, and an increased spinal fluid (CSF) protein level. A common, recurrent deletion spanning m.8470_13446 causes Kearns-Sayre syndrome; however, there are additional deletions that contribute to the syndrome. These deletions are detected in muscle.

Pearson syndrome's clinical features include sideroblastic anemia, exocrine pancreas dysfunction with symptoms in the first year of life. mtDNA deletions that cause Pearson syndrome are abundant in blood.

Chronic PEO can be the mildest of the mtDNA deletion phenotypes. This presentation is characterized by progressive ptosis, ophthalmoplegia, oropharyngeal, and proximal muscle weakness. mtDNA deletions that cause PEO are primarily detectable in muscle.

Occasionally, mtDNA deletions cause Leigh syndrome, which is characterized by psychomotor regression, abnormal brain MRI, and elevated blood and CSF lactate levels. However, other mtDNA variants may also cause Leigh syndrome. If caused by a large deletion, it is usually detectable in muscle or blood.

Large deletions can be present in only a fraction of mitochondria; a phenomenon known as heteroplasmy. Typically, the severity of disease presentation is a function of the degree of heteroplasmy. Determining the heteroplasmy of large mtDNA deletions is challenging by common clinical methods, such as next-generation sequencing. However, this droplet digital polymerase chain reaction method can obtain an accurate range of heteroplasmy levels in a variety of tissues.

Reference Values

An interpretive report will be provided.

Interpretation

The interpretation of molecular biomarker analysis includes an overview of the results and the associated diagnostic, prognostic, and therapeutic implications.

Cautions

Clinical Correlations:
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.

If testing was performed because of a clinically significant family history, it is often useful to first test an affected family member. Detection of a reportable variant in an affected family member would allow for more informative testing of at-risk individuals.

Technical Limitations:
This assay will not detect the breakpoints for large mitochondrial deletions or single nucleotide variants that cause mitochondrial disease. Therefore, the absence of a detectable variant does not rule out the possibility that an individual is affected with mitochondrial disease. This test can only detect mitochondrial DNA (mtDNA) deletions that include the *mt-ND4* or *mt-ND2* genes.

Some individuals who have a mitochondrial deletion syndrome may have a deletion that is not identified by this assay. The absence of a deletion, therefore, does not eliminate the possibility of a mitochondrial DNA deletion syndrome. For predictive testing of asymptomatic individuals, it is important to first document the presence of a deletion in an affected family member.

Of note, absence of a mitochondrial deletion does not rule out the presence of a deletion below the limits of detection of this assay (<10% heteroplasmy).

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

Clinical Reference

1. Lamont PJ, Surtees R, Woodward CE, Leonard JV, Wood NW, Harding AE. Clinical and laboratory findings in referrals for mitochondrial DNA analysis. Arch Dis Child. 1998;79(1):22-27. doi:10.1136/adc.79.1.22
2. Goldstein A, Falk MJ. Mitochondrial DNA deletion syndromes. In: Adam MP, Feldman J, Mirzaa GM, et al, eds. GeneReviews [Internet]. University of Washington, Seattle; 2003. Updated September 28, 2023. Accessed June 13, 2025. Available at www.ncbi.nlm.nih.gov/books/NBK1203/
3. Legati A, Zanetti N, Nasca A, et al. Current and new next-generation sequencing approaches to study mitochondrial DNA. J Mol Diagn. 2021;23(6), 732-741
4. McCormick EM, Lott MT, Dulik MC, et al. Specifications of the ACMG/AMP standards and guidelines for mitochondrial DNA variant interpretation. Hum Mutat. 2020;41(12):2028-2057

Performance

Method Description
This test is a droplet digital polymerase chain reaction method for determining heteroplasmy for large mitochondrial genome deletions.(Unpublished Mayo method)

PDF Report
No

Day(s) Performed
Varies

Report Available
7 to 10 days

Specimen Retention Time
Whole blood: 28 days (if available); Saliva: 30 days (if available); Extracted DNA: 3 months; Blood spots: 1 year (if available)

Performing Laboratory Location
Mayo Clinic Laboratories - Rochester Main Campus

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
81479

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
DMITO	Mitochondrial Deletion Heteroplasmy	35470-4

Test Definition: DMITO

Mitochondrial DNA Deletion Heteroplasmy, ddPCR, Varies

Result ID	Test Result Name	Result LOINC® Value
618613	Result Summary	50397-9
618614	Result	82939-0
618615	Interpretation	69047-9
618616	Additional Information	48767-8
618617	Specimen	31208-2
618618	Source	31208-2
618619	Method	85069-3
618620	Disclaimer	62364-5
618621	Released By	18771-6