# Test Definition: PMNSR

Postmortem Noonan and Related Panel

## Overview

### Useful For

Providing a comprehensive postmortem genetic evaluation in the setting of sudden cardiac death and suspicion for Noonan syndrome or related disorders

Identification of a pathogenic variant in the decedent, which may assist with risk assessment and predictive testing of at-risk family members

### Genetics Test Information

This test includes next-generation sequencing and supplemental Sanger sequencing to evaluate the *BRAF, CBL, HRAS, KRAS, MAP2K1, MAP2K2, NRAS, PTPN11, RAF1, SHOC2,* and *SOS1* genes.

### Highlights

This test is intended for use on postmortem samples (eg, formalin-fixed, paraffin-embedded [FFPE] tissue block; dried blood spot) when whole blood is not available.

This test uses next-generation sequencing to test for variants in the *BRAF, CBL, HRAS, KRAS, MAP2K1, MAP2K2, NRAS, PTPN11, RAF1, SHOC2,* and *SOS1* genes.

This test may aid in the postmortem diagnosis of Noonan syndrome, LEOPARD (lentigines, electrocardiographic conduction abnormalities, ocular hypertelorism, pulmonic stenosis, abnormal genitalia, retardation of growth, and deafness) syndrome, cardiofaciocutaneous (CFC) syndrome, Costello syndrome, or a related disorder.

Identification of a pathogenic variant may assist with familial risk assessment, screening, and genetic counseling.

### Special Instructions

- Informed Consent for Genetic Testing
- Noonan Spectrum Gene Testing Patient Information Sheet
- Informed Consent for Genetic Testing (Spanish)

### 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

### Advisory Information

This test is intended for use when EDTA whole blood is not available and formalin-fixed, paraffin-embedded (FFPE) tissue or dried blood spots are the only available samples. If EDTA whole blood is available, order NSRGP / Noonan Syndrome and Related Disorders Multi-Gene Panel, Blood.

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. Noonan Spectrum Gene Testing Patient Information Sheet **(T689) is required,** see Special Instructions. Testing may proceed without the patient information however it aids in providing a more thorough interpretation. Ordering providers are strongly encouraged to complete the form and send it with the specimen.

2. Pathology report **must** accompany specimen in order for testing to be performed. Include physician name and phone number with the specimen.

## Specimen Required

**Preferred:**

**Specimen Type:** Tissue

**Container/Tube:** Tissue block

**Collection Instructions:**Submit a formalin-fixed, paraffin-embedded tissue block.

**Additional Information:** Testing will be attempted on blocks of any age but may be canceled if adequate DNA concentration cannot be obtained.

**Specimen Stability Information:** Ambient

**Acceptable:**

**Specimen Type:** Blood spot

**Container/Tube:** Whatman FTA Classic Card or Whatman Protein Saver 903 Card

**Specimen Volume:**3-5 blood spots

**Collection Instructions:**

1. Completely fill at least 3 circles on the filter paper card

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

## 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. If not ordering electronically, complete, print, and send a Cardiovascular Test Request (T724) with the specimen.

### Specimen Minimum Volume

Tissue: See Specimen Required
Blood Spots: 3

### Reject Due To

No specimen should be rejected.

### Specimen Stability Information

| Specimen Type | Temperature | Time | Special Container |
|---|---|---|---|
| Varies | Ambient (preferred) | | |
| | Frozen | | |
| | Refrigerated | | |

## Clinical and Interpretive

### Clinical Information

Noonan syndrome (NS) is an autosomal dominant disorder of variable expressivity whose characteristic features can include short stature, congenital heart defects, characteristic facial dysmorphology, unusual chest shape, developmental delay of varying degree, cryptorchidism, and coagulation defects, among other features. In approximately 20% to 30% of cases, Noonan syndrome and related disorders are associated with hypertrophic cardiomyopathy, which may lead to sudden cardiac death. Postmortem diagnosis of Noonan syndrome or a related disorder may assist in confirmation of the cause of death, as well as risk assessment in living family members. Other heart defects associated with Noonan syndrome and related disorders include pulmonary valve stenosis (20%-50%), atrial septal defects (6%-10%), ventricular septal defects (approximately 5%), and patent ductus arteriosus (approximately 3%). Facial features, which tend to change with age, may include hypertelorism, downward-slanting eyes, epicanthal folds, and low-set and posteriorly rotated ears. Mild mental retardation is seen in up to one-third of adults.

The incidence of NS is estimated to be between 1 in 1,000 and 1 in 2,500, although subtle expression in adulthood may cause this number to be an underestimate. NS is genetically heterogeneous, with 4 genes currently associated with the majority of cases: *PTPN11, RAF1, SOS1,* and *KRAS.* Heterozygous variants in *NRAS*, *HRAS*, *BRAF*, *SHOC2*, *MAP2K1*, *MAP2K2*, and *CBL* have also been associated with a smaller percentage of NS and related phenotypes. All of these genes are involved in a common signal transduction pathway known as the Ras-mitogen-activated protein kinase (MAPK) pathway. The MAPK pathway is important for cell growth, differentiation, senescence, and death. Molecular genetic testing of all of the known genes identifies a pathogenic variant in approximately 75% of affected individuals. NS can be sporadic and due to new (de novo) variants; however, an affected parent can be recognized in 30% to 75% of families.

Some studies have shown that there is a genotype-phenotype correlation associated with NS. An analysis of a large cohort of individuals with NS has suggested that *PTPN11* variants are more likely to be found when pulmonary stenosis is present, while hypertrophic cardiomyopathy is commonly associated with *RAF1* variants, but rarely

associated with *PTPN11.*

A number of related disorders exist that have phenotypic overlap with NS and are caused by variants in the same group of genes. *PTPN11* and *RAF1* variants have been associated with LEOPARD (lentigines, electrocardiographic conduction abnormalities, ocular hypertelorism, pulmonic stenosis, abnormal genitalia, retardation of growth, and deafness) syndrome, an autosomal dominant disorder sharing several clinical features with NS. Variants in *BRAF, MAP2K1, MAP2K2,* and *KRAS* have been identified in individuals with cardiofaciocutaneous (CFC) syndrome, a condition involving congenital heart defects, cutaneous abnormalities, Noonan-like facial features, and severe psychomotor developmental delay. Costello syndrome, which is characterized by coarse facies, short stature, distinctive hand posture and appearance, severe feeding difficulty, failure to thrive, cardiac anomalies, and developmental disability has been primarily associated with variants in *HRAS*. Variation in *SHOC2* has been associated with a distinctive phenotype involving features of Noonan syndrome and loose anagen hair.

**Genes included in the Postmortem Noonan and Related Panel**

| Gene | Protein | Inheritance | Disease Association |
|---|---|---|---|
| *BRAF* | V-RAF murine sarcoma viral oncogene homolog b1 | AD | Noonan/CFC/Costello syndrome |
| *CBL* | CAS-BR-M murine ecotropic retroviral transforming sequence homolog | AD | Noonan-like syndrome disorder |
| *HRAS* | V-HA-RAS Harvey rat sarcoma viral oncogene homolog | AD | Costello syndrome |
| *KRAS* | V-KI-RAS Kirsten rat sarcoma viral oncogene homolog | AD | Noonan/CFC/Costello syndrome |
| *MAP2K1* | Mitogen-activated protein kinase, kinase 1 | AD | Noonan/CFC |
| *MAP2K2* | Mitogen-activated protein kinase, kinase 2 | AD | Noonan/CFC |
| *NRAS* | Neuroblastoma ras viral oncogene homolog | AD | Noonan syndrome |
| *PTPN11* | Protein-tyrosine phosphatase, nonreceptor-type, 11 | AD | Noonan/CFC/LEOPARD syndrome |
| *RAF1* | V-raf-1 murine leukemia viral oncogene homolog 1 | AD | Noonan/LEOPARD syndrome |
| *SHOC2* | Suppressor of clear, c. Elegans, homolog of | AD | Noonan-syndrome like with loose anagen hair |
| *SOS1* | Son of sevenless, drosophila, homolog 1 | AD | Noonan- like syndrome with loose anagen hair |

Abbreviations: autosomal dominant (AD)

## 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.

## Cautions

Sample Quality:

This test is intended for use when EDTA whole blood is not available and formalin-fixed, paraffin-embedded (FFPE) tissue or blood spots are the only available samples. DNA extracted from FFPE tissue can be degraded, which results in a higher failure rate (approximately 5%) for next-generation sequencing when compared to DNA extracted from whole blood. Due to the quality of DNA extracted from FFPE, the acceptable coverage threshold is lower than that of the equivalent blood assays. Coverage of at least 40X is expected for all regions assessed but may be adjusted on a case-by-case basis at the discretion of the laboratory director. Sanger sequencing may be used in regions that do not achieve this rate of coverage at the discretion of laboratory director. Genomic regions that are not sufficiently covered for analysis and interpretation will be indicated on the laboratory report. Sanger sequencing on DNA extracted from FFPE may also result in quality limitations when compared to testing on DNA extracted from blood.

Clinical Correlations:

Some individuals who have involvement of 1 or more of the genes on the panel may have a variant that is not identified by the methods used (eg, promoter variants, deep intronic variants). The absence of a variant, therefore, does not eliminate the possibility of Noonan syndrome or a related disorder.

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 family history of Noonan syndrome or a related disorder, it is often useful to first test an affected family member. Identification of a pathogenic variant in an affected individual would allow for more informative testing of at risk individuals.

Technical Limitations:

Next-generation sequencing may not detect all types of genetic variants. Additionally, rare variants may be present that could lead to false-negative or false-positive results. If results do not match clinical findings, consider alternative methods for analyzing these genes.

For blood spot sample type: If the patient has had an allogeneic blood or marrow transplant or a recent (ie, <6 weeks from time of sample collection) heterologous blood transfusion these results may be inaccurate due to the presence of donor DNA.

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.

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. Online Mendelian Inheritance in Man. Accessed 10/5/2017. Available at www.ncbi.nlm.nih.gov/sites/entrez?db=OMIM

2. Tartaglia M, Gelb BD, Zenker M: Noonan syndrome and clinically related disorders. Best Pract Res Clin Endocrinol Metab 2011;25(1):161-179

3. Rauen KA: Cardiofaciocutaneous Syndrome. In: Pagon RA, Adam MP, Ardinger HH, et al, eds. GeneReviews. University of Washington, Seattle. 1993-2018. 2007 Jan 18 (Updated 2016 Mar 3). Accessed 2/2018. Available at www.ncbi.nlm.nih.gov/books/NBK1186/

4. Allanson JE, Roberts AE: Noonan Syndrome. In: Pagon RA, Adam MP, Ardinger HH, et al, eds. GeneReviews. University of Washington. Seattle. 1993-2018. 2001 Nov 15 (Updated 2016 Feb 25). Accessed 2/2018. Available at www.ncbi.nlm.nih.gov/books/NBK1124/

5. Gripp KW, Lin AE: Costello Syndrome. In: Pagon RA, Adam MP, Ardinger HH, et al, eds. GeneReviews. University of Washington, Seattle. 1993-2018. 2006 Aug 29 (Updated 2012 Jan 12). Accessed 2/2018. Available at www.ncbi.nlm.nih.gov/books/NBK1507/

6. Gelb BD, Tartaglia M: Noonan Syndrome with Multiple Lentigines. In: Pagon RA, Adam MP, Ardinger HH, et al, eds. GeneReviews. University of Washington, Seattle. 1993-2018. 2007 Nov 30 (Updated 2015 May 14) Accessed 2/2018. Available at www.ncbi.nlm.nih.gov/books/NBK1383/

## 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 SureSelect Target Enrichment System. Data is analyzed with a bioinformatics software pipeline. Supplemental and/or confirmatory Sanger sequencing is performed when necessary.(Unpublished Mayo method)

The following genes are evaluated in this multigene panel: *BRAF, CBL, HRAS, KRAS, MAP2K1, MAP2K2, NRAS, PTPN11, RAF1, SHOC2,* and *SOS1.*

## PDF Report

No

## Day(s) and Time(s) Test Performed

Monday; Varies

## Analytic Time

6 weeks

### Maximum Laboratory Time

8 weeks

### Specimen Retention Time

Extracted DNA: 2 months; Client provided paraffin blocks (FFPE) and dried blood spots (DBS) will be returned to client after testing is complete.

### 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

81479

81404

81311

81405 X2

81406 X6

### LOINC® Information

| Test ID | Test Order Name | Order LOINC Value |
|---|---|---|
| PMNSR | Postmortem Noonan and Related Panel | In Process |

| Result ID | Test Result Name | Result LOINC Value |
|---|---|---|
| BA1423 | Gene(s) Evaluated | 48018-6 |
| BA1424 | Result Summary | 50397-9 |
| BA1425 | Result Details | 82939-0 |
| BA1426 | Interpretation | 69047-9 |
| BA1427 | Additional Information | 48767-8 |
| BA1428 | Method | 49549-9 |
| BA1429 | Disclaimer | 62364-5 |

| Result ID | Test Result Name | Result LOINC Value |
|---|---|---|
| BA1430 | Reviewed by | 18771-6 |