NOONAN SYNDROME AND RELATED DISORDERS

PTPN11, SOS1, RAF1, KRAS & SHOC2 GENE SEQUENCING

Noonan syndrome (OMIM 163950) is an autosomal dominant disorder due to mutations in several genes that are involved in the Ras-mitogen-activated protein kinase (RAS/MapK) pathway. Specifically, Noonan syndrome is caused by mutations in PTPN11 (OMIM 176876), SOS1 (OMIM 182530), RAF1 (OMIM 164760), and KRAS (OMIM 190070). Noonan syndrome is characterized by heart defects including hypertrophic cardiomyopathy and pulmonic valve stenosis, facial dysmorphology, short stature, chest wall deformities, and developmental delay. Noonan syndrome-like disorder with loose anagen hair (OMIM 607721) is a closely related disorder characterized by the above features as well as actively growing hair that is sparse, easy to pluck, thin, and slow-growing. This related disorder is due to mutations in SHOC2 (OMIM 602775), which is also involved in the RAS/MapK pathway.

PTPN11 and RAF1 are also associated with LEOPARD syndrome (OMIM 151100). LEOPARD syndrome is an acronym for multiple lentigines, electrocardiogram abnormalities, ocular hypertelorism, pulmonic valvular stenosis, abnormalities of genitalia, retardation of growth, and sensorineural deafness. LEOPARD syndrome is an autosomal dominant disorder, and can present much like Noonan syndrome with additional features.

Testing: Testing can be performed in tiers, moving to the next tier only if the preceding test is negative. Testing can also be performed concurrently or in any order requested. If desired, the laboratory can assist with prioritizing the order of tests if the Clinical Information Checklist is completed. The following strategy is suggested for Noonan syndrome testing:

Tier 1: Sequencing of the entire coding region of PTPN11
Tier 2: Sequencing of the entire coding region of SOS1
Tier 3: Sequencing of exons 7, 14 and 17 of RAF1
Tier 4: Sequencing of the entire coding region of KRAS
Tier 5: Sequencing of part of exon 2 for reported mutation in SHOC2

➢ Tiers 1 and 3 are suggested if an individual has findings suggestive of LEOPARD syndrome.
➢ Tier 5 is suggested if an individual has loose anagen hair.

Sequencing tests will detect point mutations, small deletions, and small insertions in the regions of the genes that are analyzed. It will not detect a partial or whole gene deletion or duplication.

For Noonan syndrome, mutations are detected in:

- PTPN11 in about 50% of affected individuals
- SOS1 in about 10% of affected individuals
- Exon 7, 14, or 17 of RAF1 in 3% to 17% of affected individuals
- KRAS in less than 5% of affected individuals
- Exon 2 of SHOC2 in individuals with Noonan-like syndrome with loose anagen hair
For LEOPARD syndrome, mutations are detected in:

- **PTPN11** in about 90% of affected individuals
- exon 7, 14, or 17 of **RAF1** in about 3% of affected individuals

A negative test does not completely rule out a diagnosis of Noonan syndrome or LEOPARD syndrome, since mutations in these five genes do not account for 100% of cases. Clinical overlap is seen between Noonan syndrome, Costello syndrome, and cardiofaciocutaneous (CFC) syndrome. Tests for Costello syndrome and CFC syndrome are also available in our laboratory and can be requested if clinically indicated.

**Turnaround time:** 7–10 days for each gene; about 3 weeks for all 5 tiers

**BILLING:** We do not bill third party payers (insurance companies) for samples received from external sources. The person or institution (e.g., Clinical Lab; Send-out Lab; Physician Office) sending the sample is responsible for full payment of the invoices within 30 days of receipt of the invoice. If the patient is on Medical assistance, please contact the lab prior to sample submission. Direct patient billing will be accepted only when a valid credit card form is received with the patient sample.

**CPT codes and cost:**

<table>
<thead>
<tr>
<th>Tier 1:</th>
<th>Tier 2:</th>
<th>Tier 3:</th>
<th>Tier 4:</th>
<th>Tier 5:</th>
</tr>
</thead>
<tbody>
<tr>
<td>83891 x 1</td>
<td>83891 x 1</td>
<td>83891 x 1</td>
<td>83891 x 1</td>
<td>83891 x 1</td>
</tr>
<tr>
<td>83898 x 14</td>
<td>83898 x 18</td>
<td>83898 x 4</td>
<td>83898 x 6</td>
<td>83893 x 1</td>
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<tr>
<td>83904 x 28</td>
<td>83904 x 42</td>
<td>83904 x 8</td>
<td>83904 x 14</td>
<td>83904 x 2</td>
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<tr>
<td>83912 x 1</td>
<td>83912 x 1</td>
<td>83912 x 1</td>
<td>83912 x 1</td>
<td>83912 x 1</td>
</tr>
</tbody>
</table>

Cost: $1400 $1500 $250 $350 $150

**Online Resources:**

**References:**

### Clinical Findings Checklist

**Costello/ CFC/ Noonan/ LEOPARD Syndrome**

(This helps the lab prioritize which gene to begin testing)

<table>
<thead>
<tr>
<th>Findings</th>
<th>Yes: select</th>
<th>No</th>
<th>Unknown</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Facial dysmorphism:</strong></td>
<td></td>
<td></td>
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<tr>
<td>□ Coarse features</td>
<td></td>
<td></td>
<td>□ Posis</td>
</tr>
<tr>
<td>□ Bitemporal narrowing</td>
<td></td>
<td></td>
<td>□ Other</td>
</tr>
<tr>
<td>□ Downslooting palpebral fissures</td>
<td></td>
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</tr>
<tr>
<td><strong>Skin/ hair findings:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>□ Deep palmar and plantar creases</td>
<td></td>
<td></td>
<td>□ Multiple lentigines</td>
</tr>
<tr>
<td>□ Fine wrinkles in palms</td>
<td></td>
<td></td>
<td>□ Multiple nevi</td>
</tr>
<tr>
<td>□ Papilloma &gt;1</td>
<td></td>
<td></td>
<td>□ Absent eyebrows</td>
</tr>
<tr>
<td>□ Hyperpigmentation</td>
<td></td>
<td></td>
<td>□ Sparse or curly hair</td>
</tr>
<tr>
<td>□ Hyperkeratosis</td>
<td></td>
<td></td>
<td>□ Thick curly hair</td>
</tr>
<tr>
<td>□ Ichthyosis</td>
<td></td>
<td></td>
<td>□ Loose anagen hair</td>
</tr>
<tr>
<td>□ Hemangioma</td>
<td></td>
<td></td>
<td>□ Other</td>
</tr>
<tr>
<td><strong>Neurologic/ sensory findings:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>□ Chiari I</td>
<td></td>
<td></td>
<td>□ Nystagmus</td>
</tr>
<tr>
<td>□ Hydrocephalus</td>
<td></td>
<td></td>
<td>□ Sensorineural hearing loss</td>
</tr>
<tr>
<td>□ Cortical atrophy</td>
<td></td>
<td></td>
<td>□ Hypotonia</td>
</tr>
<tr>
<td>□ Absence of corpus callosum</td>
<td></td>
<td></td>
<td>□ Seizures</td>
</tr>
<tr>
<td><strong>Cardiac involvement:</strong></td>
<td></td>
<td></td>
<td>□ Pulmonic valve stenosis</td>
</tr>
<tr>
<td>□ Hypertrophic cardiomyopathy</td>
<td></td>
<td></td>
<td>□ SVT</td>
</tr>
<tr>
<td>□ ASD</td>
<td></td>
<td></td>
<td>□ Other</td>
</tr>
<tr>
<td>□ VSD</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>□ Mitral valve dysplasia</td>
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<tr>
<td><strong>Feeding problems:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>□ Very difficult, no feeding tube</td>
<td></td>
<td></td>
<td>□ Short stature (&lt;2nd percentile)</td>
</tr>
<tr>
<td>□ Had feeding tube, not needed any more</td>
<td></td>
<td></td>
<td>□ Short neck</td>
</tr>
<tr>
<td>□ Still has feeding tube</td>
<td></td>
<td></td>
<td>□ Broad and webbed neck</td>
</tr>
<tr>
<td>□ Other</td>
<td></td>
<td></td>
<td>□ Macrolephaly</td>
</tr>
<tr>
<td>□ Other</td>
<td></td>
<td></td>
<td>□ Other</td>
</tr>
<tr>
<td><strong>Growth/ skeletal findings:</strong></td>
<td></td>
<td></td>
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<tr>
<td>□ Joint laxity</td>
<td></td>
<td></td>
<td>□ Short stature (&lt;2nd percentile)</td>
</tr>
<tr>
<td>□ Ulnar deviation of wrist and fingers</td>
<td></td>
<td></td>
<td>□ Short neck</td>
</tr>
<tr>
<td>□ Pectus</td>
<td></td>
<td></td>
<td>□ Broad and webbed neck</td>
</tr>
<tr>
<td>□ Kyphoscoliosis</td>
<td></td>
<td></td>
<td>□ Macrolephaly</td>
</tr>
<tr>
<td>□ Other</td>
<td></td>
<td></td>
<td>□ Other</td>
</tr>
<tr>
<td><strong>Malignancy:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>□ Rhabdomyosarcoma</td>
<td></td>
<td></td>
<td>□ ALL</td>
</tr>
<tr>
<td>□ Neuroblastoma</td>
<td></td>
<td></td>
<td>□ JMML</td>
</tr>
<tr>
<td>□ Bladder carcinoma</td>
<td></td>
<td></td>
<td>□ Myelodysplastic syndrome</td>
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<tr>
<td>□ AML</td>
<td></td>
<td></td>
<td>□ Other</td>
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<tr>
<td>□ Other</td>
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<td></td>
<td>□ Other</td>
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<tr>
<td><strong>Cognitive delay:</strong></td>
<td></td>
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<tr>
<td>□ Developmental delay</td>
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<td>□ Other</td>
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<tr>
<td>□ Learning disabilities</td>
<td></td>
<td></td>
<td>□ Other</td>
</tr>
<tr>
<td>□ Mild to moderate mental retardation</td>
<td></td>
<td></td>
<td>□ Other</td>
</tr>
<tr>
<td>□ Moderate to profound mental retardation</td>
<td></td>
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<td>□ Other</td>
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<tr>
<td><strong>Other:</strong></td>
<td></td>
<td></td>
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<tr>
<td>□ Polyhydramnios in utero</td>
<td></td>
<td></td>
<td>□ Lymphedema</td>
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<tr>
<td>□ Cryptorchidism</td>
<td></td>
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<td>□ Coagulation defects</td>
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<tr>
<td>□ Renal anomalies</td>
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<td></td>
<td>□ Other</td>
</tr>
<tr>
<td>□ Disordered/ delayed puberty</td>
<td></td>
<td></td>
<td>□ Other</td>
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</tbody>
</table>

**Other medical problems not listed above:**

________________________________________________________

________________________________________________________
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Name of Physician/Lab receiving Fax: ____________________________________________

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Signature: ___________________________ Date: ______________

Title: ____________________________________________

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If you have any questions regarding this form please contact Susan Kirwin, Assistant Director of the Molecular Diagnostics Laboratory, at 302.651.6777.

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Credit Card Billing Information

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<th>American Express</th>
<th>Discover</th>
<th>Other: _____________</th>
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<tbody>
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<td>Credit Card Number:</td>
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<tr>
<td>Card holder address:</td>
<td>Expiration Date:</td>
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<tr>
<td>Card holder phone:</td>
<td>Security Code:</td>
<td>Home:</td>
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<td>(on back of card)</td>
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<td>Work:</td>
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<tr>
<td>Card holder signature:</td>
<td>Authorized payment amount:</td>
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</tbody>
</table>

For Direct Patient Billing

Prepayment for the testing services is required prior to beginning our testing. Please complete this form and include this paperwork with the shipment of the patient sample.

Billing questions can be addressed to: Denise Axsmith
Senior Budget/Financial Analyst
Nemours/A.I. duPont Hospital for Children
daxsmith@nemours.org
Phone: 302.651.6802
Fax: 302.651.6881