Spinal Muscular Atrophy (SMA) testing
including: SMA Carrier Testing – or – SMA Diagnostic Testing

TEST DESCRIPTION:
Spinal Muscular Atrophy (SMA) is a leading cause of progressive muscle weakness and atrophy with an approximate incidence of 1 in 10,000 births.\(^1\) It is inherited in an autosomal recessive manner and can present with a spectrum of severity and onset that spans five different types (0-IV).\(^2\)

SMA testing offered through our laboratory is capable of detecting both \(SMN1\) and \(SMN2\) copy number, together with common polymorphisms indicative of a haplotype that correlates with an increased risk for carrier status. This testing can be ordered as a screening test for the purpose of determining carrier status or as a diagnostic test.

- **Carrier testing** is performed to determine the genetic status of asymptomatic individuals with respect to this condition. Carriers of SMA are not expected to be symptomatic. This testing is most often performed in a prenatal or preconception testing in order to provide information about recurrence risks, but may also be indicated in parents of an affected individual.
- **Diagnostic testing** is performed most often in a pediatric or adult patient care setting for the purpose of determining whether or not an individual is affected with SMA. Confirmation of a diagnosis can have important implications for medical management; testing can also provide additional prognostic information in an individual for whom the diagnosis was made by a different methodology.

TEST DETAILS:
Our Spinal Muscular Atrophy (SMA) testing includes both \(SMN1\) and \(SMN2\) analysis, as well interrogation of two common polymorphisms (g.27134T>G and g.27706-27707delAT).

- Both \(SMN1\) and \(SMN2\) copy number is assessed by Multiplex Ligation-dependent Probe Amplification (MLPA), which detects the quantitative dosage of exons 7 and 8 of \(SMN1\) and exon 7 of \(SMN2\). Gene dosage ratios are calculated relative to the average 18 reference loci expressed as copy numbers.
- This methodology also detects the presence or absence of the two common polymorphisms indicated above. The presence of the T>G variant is consistent with presumed carrier status in the Ashkenazi Jewish and Asian populations and with an increased carrier risk in the Caucasian, African American, and Hispanic populations.\(^2\)

The rate of detection for this test varies based on ethnicity. For carrier screening, residual risk is reduced when two copies of \(SMN1\) are detected. The residual risk may be further reduced in the event that three copies of \(SMN1\) are detected in an individual. The presence of the common polymorphisms is suggestive of silent carrier status with a potentially significant increase in the risk for that individual to be a carrier.

<table>
<thead>
<tr>
<th>Ethnicity</th>
<th>Pre-test carrier risk</th>
<th>Rate of carrier detection</th>
<th>Residual risk after a negative result</th>
<th>Residual risk with 2 copies of (SMN1) and positive result for g.27134T&gt;G</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ashkenazi Jewish</td>
<td>1/41</td>
<td>94%</td>
<td>1/580</td>
<td>~1*</td>
</tr>
<tr>
<td>Asian</td>
<td>1/53</td>
<td>93%</td>
<td>1/702</td>
<td>~1*</td>
</tr>
<tr>
<td>Caucasian</td>
<td>1/35</td>
<td>95%</td>
<td>1/769</td>
<td>1/29</td>
</tr>
<tr>
<td>African American</td>
<td>1/66</td>
<td>71%</td>
<td>1/396</td>
<td>1/34</td>
</tr>
<tr>
<td>Hispanic</td>
<td>1/117</td>
<td>91%</td>
<td>1/1,762</td>
<td>1/140</td>
</tr>
</tbody>
</table>

Table adapted from Luo et al. 2014.\(^3\)
In the case of diagnostic testing, carrier status may also be reported. If there is strong suspicion for SMA in an individual with a single copy of \( \text{SMN1} \) detected, further studies such as gene sequencing may be warranted.

\( \text{SMN2} \) copy number will only be reported for individuals with homozygous deletion of \( \text{SMN1} \), consistent with a diagnosis of SMA. Information about \( \text{SMN2} \) copy number is available upon request for individuals with a result that is consistent with heterozygous deletion or other high-risk carrier status.

**COMPLEMENTARY TESTING:**
Additional testing can be considered depending on the indication for testing and is available as concurrent or reflex testing options.

**Carrier Testing**
- **Cystic fibrosis (CF) carrier testing** is also available and can be paired with SMA carrier testing. Both CF and SMA carrier testing are recommended by ACOG to be offered to all couples who are pregnant or considering pregnancy.\(^3\) CF carrier testing is performed and reported by Nebraska Medicine Molecular Diagnostic Laboratory but can be ordered on the Human Genetics Laboratory test request form.
- **Fragile X carrier testing** can also be considered for all couples who are pregnant or considering pregnancy, and is especially indicated for women with a family history of autism or cognitive impairment. Fragile X testing is performed and reported by Nebraska Medicine Molecular Diagnostic Laboratory but can be ordered on the Human Genetics Laboratory test request form.
- **Chromosome analysis** can be considered in the prenatal care setting for couples with a history of recurrent miscarriage or for individuals with a family history of chromosome abnormality or unexplained neurodevelopmental disorder.
- Other gene-specific testing can be offered as indicated by review of the family history.

**Diagnostic Testing**
- **Chromosome analysis** or **SNP array analysis** may be warranted depending on the patient indication for testing.
- Additional gene-specific testing may be warranted depending on patient findings. Please contact the laboratory for assistance in curation of a customized gene panel if desired.

**ADVANTAGES:**
- Detection of dosage for \( \text{SMN1} \) exons 7 and 8 is considered standard testing for SMA. Analysis performed at the Human Genetics Laboratory also offers \( \text{SMN2} \) copy number quantification, which can provide important prognostic information for affected individuals.\(^2\)
- Two sequence-level variants are also detected through this methodology, the presence of which is suggestive of silent carrier status.\(^3\) Although residual risk may vary based on ethnicity, this can provide a more accurate carrier screen than the standard \( \text{SMN1} \) analysis alone.
- SMA testing in the diagnostic setting is important for medical decision making. Individuals with a confirmation of a genetic diagnosis may be eligible for additional targeted therapies and treatments.\(^2,4\)
- Prenatal diagnosis may be available for couples when both individuals are confirmed to be a carrier of SMA (prenatal testing for SMA on a fetal specimen is not available at the Human Genetics Laboratory).

**LIMITATIONS:**
- Testing is unable to detect if two copies of \( \text{SMN1} \) are on the same chromosome (a 2+0 genotype, considered silent carrier status); however, an increased risk of such status can be estimated through the interrogation of two common polymorphisms.
- This assay cannot rule out the possibility of disease-causing variants at other loci not evaluated or sequence-level variants in \( \text{SMN1} \) other than the two polymorphisms indicated above.
- Although unlikely, if aberrant copy numbers exist for any of the reference loci, the analysis may be compromised.
- This test has been validated on peripheral blood specimens only. SMA testing on other sample types (e.g. buccal mucosa, amniocytes) is not available at the Human Genetics Laboratory.
INDICATIONS FOR CARRIER TESTING:

- Family history of SMA or an individual with features suggestive of SMA
- Any couple currently pregnant or considering a pregnancy (recommended by both ACOG and ACMG\(^5\)\(^6\))

INDICATIONS FOR DIAGNOSTIC TESTING:

- Motor delay and/or progressive loss of motor skills
- Proximal muscle weakness
- Hyptonia
- Decreased reflexes
- Tongue fasciculations
- Lack of movement in infancy
- Restrictive lung disease or recurrent aspirations
- Joint contractures and/or scoliosis in childhood

SPECIMEN COLLECTION & TRANSPORT:

Complimentary test kits are available upon request, but are not required.

SAMPLE TYPE and REQUIREMENTS:

- blood:
  - 2-5 ml whole blood in an EDTA tube (purple top) for SMA testing or CF testing
  - If ordering testing for both SMA and CF, one tube is sufficient

SHIPPING:

- Maintain and ship samples at room temperature.
- Coordinate transport for sample to be received in our laboratory within 24-48 hours of collection.
  - LOCAL: Call 402-559-5070 (option 1)
  - OUT OF AREA: Prior to shipment, please fax the completed test request form to 402-559-7248, including the FedEx\(^\text{®}\) airbill tracking number.
    - Saturday delivery MUST be checked when sending FedEx\(^\text{®}\) on Friday.
    - Please include Internal Billing Reference # 3155070600 on the FedEx\(^\text{®}\) airbill.
    - Ship To: Human Genetics Laboratory – Zip 5440
      UNMC Shipping & Receiving Dock
      601 S. Saddle Creek Road
      Omaha, NE 68106

FORMS FOR TESTING: The following forms can be downloaded via our website.

- Required: Postnatal Test Request Form
- Optional: Informed Consent for Genetic Testing

POTENTIAL TEST RESULTS:

- Normal: Two or more copies of \(SMN1\) are detected.
- Carrier: One copy of \(SMN1\) exon 7 is detected, consistent with status as a carrier of SMA.
- Silent Carrier Status: Two copies of \(SMN1\) are detected, but the presence of one or both sequence-level variants is suggestive of silent carrier status.
- Abnormal: No copies of \(SMN1\) exon 7 are detected, consistent with a diagnosis of SMA.

TURNAROUND TIME: Results are typically available in 2 - 3 weeks.
BILLING: Our laboratory offers patient/self-pay, insurance (including Medicare/Medicaid), and client/institution billing options. Verifying coverage requirements or obtaining preauthorization PRIOR TO OR AT THE TIME OF SPECIMEN COLLECTION is often necessary. We provide preauthorization services upon request by calling 402-559-5070 (option 3); the following form is helpful for obtaining the information required by insurance providers and can be downloaded via our website.

- Insurance Preauthorization Request

In some circumstances, a test may be warranted even though insurance coverage is denied or not guaranteed. For these situations, we request the following form be signed by the patient and submitted with the sample. This helps inform patients of their potential financial responsibility, should the costs of genetic testing not be paid by their insurance provider.

- Advanced Beneficiary Notice of Noncoverage (ABN) – required when billing Medicare

CPT CODES:

- SMA: 81329

PRICING: For current costs contact the laboratory billing staff at 402-559-5070 (option 3).

REFERENCES:

updated 7/2019