Spinal Muscular Atrophy

Indications for Ordering

- Diagnostic testing to confirm a suspected diagnosis of spinal muscular atrophy (SMA)
- Quantify SMN2 copy number for treatment purposes
- Prenatal or preconception carrier screening for SMA in the general population
- Carrier screening for reproductive partner of known SMA carrier
- Carrier screening for parents of a child with a deletion of the SMN1 gene or other family history of SMA

Test Description

Multiplex ligation-dependent probe amplification (MLPA) to determine copy number for the SMN1 and SMN2 genes

Tests to Consider

Primary tests
Spinal Muscular Atrophy (SMA) Copy Number Analysis 2013436
- Diagnostic or carrier testing for SMA
  - Although relevant only for affected individuals, SMN2 copy number will be reported for all patients
  - Chromosomal phase cannot be determined
  - Test also includes 2 SMN1 variants that are part of a haplotype associated with SMN1 duplication in silent carriers, especially in Ashkenazi Jewish and Asian populations
    - c.*3+80T>G (rs143838139), g.27134T>G
    - c.*211_*21del (rs200800214), g.27706-27707delAT
    - Relevant only in the context of carrier screening, but will be reported for all patients as present or not present

Spinal Muscular Atrophy (SMA) Copy Number Analysis, Fetal 2013444
- Prenatal diagnostic testing for SMA when both parents carry a deletion of SMN1 or have a previous child with SMA caused by a deletion of SMN1
  - SMN2 copy number will be reported

Related Test

Genetic Carrier Screen (CF, FXS, and SMA) with Reflex to Methylation 3000258
- Screen for genetic variants that indicate carrier status for cystic fibrosis (CF), fragile X syndrome (FXS), and spinal muscular atrophy (SMA) in pregnant couples or those planning a pregnancy
- Do not use for diagnostic testing in patients with symptoms of CF, FXS, or SMA

Disease Overview

Incidence – ~1/10,000 live births in the U.S.
- Carrier rate ~1/50 overall in the U.S.; varies by ethnicity
  - Caucasians – 1/35
  - Ashkenazi Jews – 1/41
  - Asians – 1/53
  - African Americans – 1/66
  - Hispanics – 1/117

Symptoms

- Progressive muscle weakness due to degeneration of lower motor neurons
  - Clinical findings of affected individuals fall on a spectrum
  - Most common symptoms
    - Difficulty breathing, swallowing, and walking
  - SMA subtypes are distinguished by age of onset and severity for purposes of prognosis and management
    - SMA 0 – prenatal onset
      - Most severe form, survival is typically <6 months
    - SMA 1 – onset at 0-6 months
      - Severe muscle weakness, survival is typically <2 years
    - SMA 2 – onset at 6-12 months
      - Child usually cannot walk without assistance
    - SMA 3 – onset after 12 months
      - Milder muscle weakness, child usually can walk and stand without assistance
    - SMA 4 – adult onset
      - Mild muscle weakness, normal life span
**Diagnostic testing**

- Diagnosis is based on clinical findings and molecular genetic testing
  - Electromyography (EMG), nerve conduction velocities (NCV), and muscle/nerve histology may aid in diagnosis
- 95-98% of individuals with SMA have a homozygous loss of SMN1 (0 copies of SMN1)
- 2-5% of individuals with SMA have loss of SMN1 on 1 chromosome and a pathogenic sequence variant in the remaining copy of SMN1 (not detected by this test)
- Not possible to definitively predict clinical subtype based on genotype
  - Higher SMN2 copy number may correlate with milder disease severity in affected individuals

**Carrier testing**

- Presence of ≥2 copies of SMN1 usually indicates patient is not a carrier, although residual carrier risk exists
  - Test is unable to determine if SMN1 copies are on the same or opposite chromosome
- ~3-4% of general population has both copies of SMN1 on the same chromosome (also known as SMN1 duplication)
  - If paired with SMN1 loss (0 copies) on the opposite chromosome, these individuals are “silent carriers” or “2+0 carriers”
- ≥2 copies of SMN1 on the same chromosome is rare but more frequent in certain populations
  - African Americans, Ashkenazi Jews
- 2 variants which are part of a haplotype associated with SMN1 duplication are tested
  - Presence of 2 SMN1 copies plus the variants suggest increased risk that the individual is a silent carrier but is not definitive
  - Likely silent carrier if patient is Ashkenazi Jewish or Asian

**Pathophysiology**

- SMA is caused by low levels of survival motor neuron (SMN) protein essential for motor neurons
- Majority of full-length SMN protein production comes from SMN1 gene
- Some full-length SMN protein production comes from SMN2 gene
  - SMN2 differs very little from SMN1
    - Does not contain exon 7
      - Alters mRNA splicing and protein production
    - Usually multiple copies of SMN2 per chromosome
      - Increased full-length SMN protein production from SMN2 may partially compensate for SMN protein production missing due to altered SMN1 gene
- Spinraza (nusinersen) is a FDA-approved drug that can be used to treat SMA by increasing the amount of full-length SMA protein produced from SMN2

**Genetics**

**Genes** – SMN1, SMN2

**Inheritance** – autosomal recessive

- 98% of SMA is caused by abnormality in both copies of SMN1 gene

**De novo rate** – 2%, usually paternal in origin

**Variants tested/reported**

- SMN1 copy number
  - Does not report mechanism (gene deletion vs gene conversion)
  - Variants tested for haplotype associated with SMN1 duplication
    - c.*2+80T>G (rs143838139)
    - c.*211_*212del (rs200800214)
- SMN2 copy number will be reported for all individuals

**Test interpretation**

**Sensitivity/specificity**

- Clinical sensitivity for diagnostic testing (Hendrickson, 2009; Luo, 2014; Prior, 2013)
  - 95-98% of individuals with SMA have 0 copies of SMN1
  - 2-5% of individuals with SMA have 1 copy of SMN1 plus a pathogenic sequence variant
- Detection rate for carrier screening
  - Caucasians – 95%
  - Ashkenazi Jews – 94%
  - Asians – 93%
  - Hispanics – 91%
  - African Americans – 71%
- Analytical sensitivity/specificity – 99%

**Results**

- Diagnostic test results
  - 0 copies of SMN1 detected
    - Consistent with diagnosis of SMA
  - 1 copy of SMN1 detected
    - May have SMA if an undetected pathogenic sequence variant is present
    - At least a carrier for SMA
  - 2 copies of SMN1 detected
    - Not affected with SMA
- Carrier screening results
  - 1 copy of SMN1 detected
    - Individual is a carrier for SMA
  - 2 or more copies of SMN1 detected
    - Carrier risk is reduced but not eliminated
  - 2 copies of SMN1 detected and positive for targeted variant(s)
    - Normal copy number but increased risk to be a silent carrier (may have both SMN1 copies on same chromosome, 0 copies on the other chromosome)
    - Not definitive
Limitations

- Diagnostic errors can occur due to rare sequence variations
- Single base pair substitutions, small deletions/duplications, regulatory region mutations, and deep intronic mutations will not be detected
- Test is unable to determine
  - Whether SMN1 copies are on the same or opposite chromosomes
    - >1 copy of SMN1 on each chromosome (not a carrier) indistinguishable from >2 copies of SMN1 on 1 chromosome and 0 copies on the opposite chromosome (silent carrier)
  - Whether SMN1 and SMN2 copies are on the same or opposite chromosomes

References