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# CLSI NBS13™

## Newborn Screening for Spinal Muscular Atrophy

CLSI NBS13 describes a newborn screening system for detecting spinal muscular atrophy (SMA). It discusses both *SMN1* exon 7 deletion testing and *SMN2* copy number analysis performed on newborn dried blood spot specimens, as well as screening strategies for identifying newborns at increased risk for SMA.

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A guideline for global application developed through the Clinical and Laboratory Standards Institute consensus process.

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### Abstract

Clinical and Laboratory Standards Institute NBS13—*Newborn Screening for Spinal Muscular Atrophy* describes newborn screening (NBS) laboratory tests and screening strategies used to identify newborns at increased risk of developing spinal muscular atrophy (SMA). SMA is a serious condition that causes weakness, motor decline, and death in otherwise cognitively normal children. Early detection and intervention are critical to improving the strength and survival of newborns with SMA, allowing them to live healthier and more productive lives. SMA is caused by loss of function of the survival of motor neuron (SMN) gene, with about 95% of SMA cases characterized by a homozygous deletion in *SMN1* exon 7, leading to deficiency of the SMN protein and degeneration of motor neurons followed by progressive muscle weakness. The severity of the disease is typically inversely correlated to the number of *SMN2* copies. CLSI NBS13 describes the various methods used for *SMN1* exon 7 deletion testing and *SMN2* copy number analysis, validation of assays, integrating SMA NBS into NBS laboratory operations, interpreting and reporting screening results, as well as short-term and long-term follow-up.

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## Foreword

Spinal muscular atrophy (SMA) is a disorder of progressive weakness associated with loss of lower motor neurons (LMNs), which are the nerve cells that originate in the anterior horn of the spinal cord. Although rare, SMA is one of the more common inherited causes of childhood death. SMA is caused by deficiency of the survival of motor neuron (SMN) protein, which is usually due to loss of function of *SMN1* on chromosome 5q. SMN protein is not absent, however, because of the presence of a nearly identical pseudogene, *SMN2*, that can produce small amounts of SMN protein. Historically, SMA has been categorized into a variety of types, largely based on how much residual SMN protein is produced by *SMN2*. Children with SMA have weakness that progresses as their LMNs continue to die. It is estimated that in SMA type 1, the severe infantile form of SMA, up to 90% of LMNs die by age 6 months, leaving the infant unable to sit and unable to breathe without chronic ventilatory support. Since 2016, three effective disease-modifying treatments (DMTs) for SMA have been approved, all of which effectively increase the amount of SMN protein and, if given early enough, prevent LMN degeneration. With these treatments, children with SMA who are treated before any LMN loss can make ongoing and normal motor gains, rather than motor declines.

The commercial availability of the first DMT, nusinersen, at the end of 2016 meant that if treated early enough, children with SMA type 1 could make typical motor gains in infancy without needing respiratory support. The remarkable changes in outcomes seen in the clinical trials of nusinersen helped SMA receive approval for inclusion on the United States' Federal Recommended Uniform Screening Panel in 2018. Since then, two additional DMTs for SMA have been approved by the US Food and Drug Administration and SMA newborn screening (NBS) has been introduced in nearly every US state, as well as internationally. The notable speed of implementation of SMA NBS in the United States was helped by the earlier introduction of severe combined immunodeficiency (SCID) which also uses DNA-based technology rather than determination of an analyte concentration that is more typical of NBS for other diseases.

**NOTE:** The content of CLSI NBS13 is supported by the CLSI consensus process and does not necessarily reflect the views of any single individual or organization.

### KEY WORDS

dried blood spots

lower motor neurons

newborn screening

real-time quantitative  
polymerase chain reaction

*SMN1* homozygous exon 7

*SMN2* copy number

spinal muscular atrophy

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# Chapter ①

## Introduction

# Newborn Screening for Spinal Muscular Atrophy

## 1 Introduction

### 1.1 Scope

CLSI NBS13 discusses the detection of 5q spinal muscular atrophy (SMA) by population-based newborn dried blood spot (DBS) screening and preanalytical, analytical, and postanalytical aspects of newborn screening (NBS) for SMA. It focuses on analytical methods to detect the homozygous absence of *SMN1* exon 7, which accounts for approximately 95% of SMA cases, as well as methods for *SMN2* copy number analysis. CLSI NBS13 describes:

- Background information on the biological and clinical features of SMA, and the value of the *SMN2* copy number in predicting disease severity
- Preanalytical considerations, including population education and issues of consent before screening
- Analytical methodologies for detecting the homozygous absence of *SMN1* exon 7, including:
  - Real-time quantitative PCR (qPCR)
  - High-resolution melting analysis (HRMA)
  - Multiplex ligation-dependent probe amplification (MLPA)
  - Digital PCR (dPCR)/droplet digital PCR (ddPCR)
  - Capillary electrophoresis
  - Mass spectrometry
- Analytical methodologies for *SMN2* copy number analysis, including real-time qPCR, dPCR, and capillary electrophoresis
- Screening strategies and laboratory screening algorithms, cutoff value determinations, case definition, and risk assessment for NBS programs to consider when implementing SMA NBS
- Postanalytical short-term follow-up (STFU) and long-term follow-up (LTFU) procedures, including case tracking, diagnostic testing needed to confirm an SMA diagnosis, and recent treatments available for SMA

The intended users of CLSI NBS13 include NBS laboratories, follow-up and program personnel, public health program administrators, diagnostic medical laboratories, SMA treatment centers, health care providers (HCPs; eg, primary care providers, neonatologists, pediatricians, neurologists), regulatory agencies, public health policy makers, and manufacturers of instruments, reagents, and related products used for NBS.

CLSI NBS13 does not include:

- Details of carrier screening
- Details of confirmatory diagnostic laboratory testing
- Details of clinical treatments and monitoring
- Comparative cost information