

# NBS03

## Newborn Screening for Preterm, Low Birth Weight, and Sick Newborns

This guideline describes the recommended protocols for screening preterm, low birth weight, and sick newborns for hearing loss, critical congenital heart defects, and diseases detectable through newborn dried blood spot screening.

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

---

## Newborn Screening for Preterm, Low Birth Weight, and Sick Newborns

Julie Luedtke, BS  
Dianne R. Webster, PhD, FHGSA  
Derek K. Bair, MD, FAAP  
Frank Bloomfield, BSc (Hons), MBChB, MRCP (UK), FRACP, PhD  
Peter J. Borandi, MBA  
Debra Freedenberg, MD, PhD, FFACMG  
Amy Gaviglio, MS, CGC  
Patrick V. Hopkins, BS  
Joseph J. Orsini, PhD  
Walter W. Reichert  
Daniel T. Robinson, MD  
Deborah Rodriguez, RN, MPH, CPH

### Abstract

Clinical and Laboratory Standards Institute guideline NBS03—*Newborn Screening for Preterm, Low Birth Weight, and Sick Newborns* describes newborn screening (NBS) of preterm, low birth weight (LBW), and sick newborns worldwide to detect treatable diseases before physical damage can occur. In developed countries, treatment advances have improved survival rates, making early disease detection by NBS even more important. The physiological states associated with preterm, LBW, and sick newborns and the treatments received directly affect the reliability of results for many diseases screened in public health by newborn dried blood spot screening, newborn hearing screening, and critical congenital heart disease (CCHD) screening. This guideline describes the effects of maternal and newborn conditions, as well as treatments given to newborns, that may affect NBS results. This guideline also provides the rationale for recommended screening intervals designed to minimize the risk of missing or delaying a diagnosis in an affected newborn. This guideline is intended for use by those involved in any aspect of NBS specimen collection, hearing screening, CCHD screening, and follow-up, including health care providers, public health professionals, and others concerned with newborn health and welfare.

Clinical and Laboratory Standards Institute (CLSI). *Newborn Screening for Preterm, Low Birth Weight, and Sick Newborns*. 2nd ed. CLSI guideline NBS03 (ISBN 978-1-68440-052-2 [Print]; ISBN 978-1-68440-053-9 [Electronic]). Clinical and Laboratory Standards Institute, 950 West Valley Road, Suite 2500, Wayne, Pennsylvania 19087 USA, 2019.

The Clinical and Laboratory Standards Institute consensus process, which is the mechanism for moving a document through two or more levels of review by the health care community, is an ongoing process. Users should expect revised editions of any given document. Because rapid changes in technology may affect the procedures, methods, and protocols in a standard or guideline, users should replace outdated editions with the current editions of CLSI documents. Current editions are listed in the CLSI catalog and posted on our website at [www.clsi.org](http://www.clsi.org). If you or your organization is not a member and would like to become one, or to request a copy of the catalog, contact us at: Telephone: +1.610.688.0100; Fax: +1.610.688.0700; E-Mail: [customerservice@clsi.org](mailto:customerservice@clsi.org); Website: [www.clsi.org](http://www.clsi.org).



CLINICAL AND  
LABORATORY  
STANDARDS  
INSTITUTE®

Copyright ©2019 Clinical and Laboratory Standards Institute. Except as stated below, any reproduction of content from a CLSI copyrighted standard, guideline, derivative product, or other material requires express written consent from CLSI. All rights reserved. Interested parties may send permission requests to [permissions@clsi.org](mailto:permissions@clsi.org).

CLSI hereby grants permission to each individual member or purchaser to make a single reproduction of this publication for use in its laboratory procedures manual at a single site. To request permission to use this publication in any other manner, e-mail [permissions@clsi.org](mailto:permissions@clsi.org).

### **Suggested Citation**

CLSI. *Newborn Screening for Preterm, Low Birth Weight, and Sick Newborns*. 2nd ed. CLSI guideline NBS03. Wayne, PA: Clinical and Laboratory Standards Institute; 2019.

### **Previous Editions:**

December 2008, October 2009

SAMPLE

ISBN 978-1-68440-052-2 (Print)  
ISBN 978-1-68440-053-9 (Electronic)  
ISSN 1558-6502 (Print)  
ISSN 2162-2914 (Electronic)

Volume 39, Number 9

## Contents

Abstract.....	i
Committee Membership.....	iii
Foreword.....	vii
Chapter 1: Introduction.....	1
1.1 Scope.....	1
1.2 Background.....	2
1.3 Standard Precautions.....	4
1.4 Terminology.....	4
Chapter 2: Overview of Newborn Screening for Preterm, Low Birth Weight, and Sick Newborns.....	9
Chapter 3: Preanalytical Considerations for Dried Blood Spot Screening of Preterm, Low Birth Weight, and Sick Newborns.....	11
3.1 Specimen Collection Issues.....	11
3.2 Recommendations for Dried Blood Spot Specimen Collection Timing.....	12
3.3 Alternative Recommendations for Dried Blood Spot Specimen Collection Timing.....	17
3.4 Special Considerations for Dried Blood Spot Specimen Collection Timing.....	19
Chapter 4: Analytical Considerations for Newborn Screening Results Interpretation.....	21
4.1 Effects of Maternal and Newborn Conditions and Treatments on Newborn Screening Test Results.....	21
4.2 Effects of Preterm, Low Birth Weight, or Sick Newborn Conditions and Treatments on Newborn Dried Blood Spot Screening Test Results.....	24
4.3 Effects of Factors Associated With Special Care Baby Unit and/or Neonatal Intensive Care Unit Populations on Newborn Point-of-Care Screening Test Results.....	29
Chapter 5: Postanalytical Activities for Newborn Screening Test Results Follow-up.....	33
5.1 Newborn Screening Program Follow-up of Positive Newborn Screening Test Results.....	33
5.2 Roles and Responsibilities for Newborn Screening Follow-up After Admission to Special Care Baby Unit and/or Neonatal Intensive Care Unit.....	33
Chapter 6: Newborn Screening System Quality Assurance, Education, and Research.....	35
6.1 Newborn Screening System Evaluation, Quality Assurance, and Performance Indicators.....	35
6.2 Newborn Screening Program Education Opportunities.....	36
6.3 Newborn Screening Research Opportunities.....	36
Chapter 7: Conclusion.....	38
Chapter 8: Supplemental Information.....	38
References.....	39
Additional Resource.....	44
Appendix A. Summary of Maternal Factors Affecting Newborn Screening Test Results.....	45
Appendix B. Summary of Newborn Factors Affecting Newborn Screening Test Results.....	47

**Contents (Continued)**

Appendix C. Factors Affecting Newborn Screening Test Results for Preterm, Low Birth Weight, and Sick Newborns .....51

Appendix D. Factors Affecting Newborn Screening by Tandem Mass Spectrometry for Preterm, Low Birth Weight, and Sick Newborns .....58

Appendix E. Acylcarnitines Measured by Tandem Mass Spectrometry .....67

The Quality Management System Approach .....68

Related CLSI Reference Materials .....70

SAMPLE

## Foreword

This guideline focuses on unique issues affecting newborn screening (NBS) for the population of preterm, low birth weight (LBW), or sick newborns. Worldwide, of the approximately 135 million births per year, about 15 million are preterm.<sup>1</sup> As many as 30 million LBW babies are born annually (23.8% of all births),<sup>2</sup> and approximately 7.9 million newborns are born with serious birth defects, of whom approximately 303 000 do not survive the newborn period.<sup>3</sup> In developed and developing countries, babies with the highest medical needs are cared for in neonatal intensive care units (NICUs), while those who have medical needs above the level of care needed by healthy newborns are cared for in special care baby units (SCBUs). For the purposes of this guideline, SCBU/NICU indicates facilities providing care above the level provided for newborns rooming with the mother or in well-baby nurseries. The SCBU/NICU population is the focus of this guideline.

Preterm birth is defined as <37 completed weeks (259 days) of gestation, and LBW is defined as <2500 g.<sup>4</sup> In countries where a higher percentage of newborns weigh <2500 g and may or may not be considered candidates for specialized care, these guidelines may not apply. Although preterm birth rates have recently declined in some countries, survivability at a lower gestational age has increased because of advances in medical care. Since the first edition of this guideline was published, earlier administration (within the first hours after birth) of medical and/or nutritional interventions affecting NBS results has become more common. The number and types of diseases screened also continue to grow, increasing complexity and making it even more challenging to ensure NBS program protocols provide sufficiently reliable results for this population.

Each disease has an optimal screening window, in which there is the greatest chance for diagnosis and treatment before symptoms or permanent damage occur. The ideal screening window is different for preterm and LBW newborns vs term newborns for several diseases, particularly cystic fibrosis, congenital hypothyroidism, congenital adrenal hyperplasia, severe combined immunodeficiency (SCID), some lysosomal storage disorders (LSDs), and critical congenital heart disease (CCHD). Screening windows may be different for some assays used to screen this population, owing to poor specificity (disproportionately high number of false-positive screen results) or poor sensitivity (unacceptably high number of false-negative screen results).

When this guideline was revised, experts in neonatology, NBS, genetics, and pediatric subspecialties reviewed the significance and duration of potential interferences, providing recommendations for timing and number of specimens needed to detect diseases on NBS panels. There remains a need for additional published research on screening complexity in this population, eg, answers are still needed about the duration of some treatments' effects on various screening results.

This guideline provides recommendations and reference information for NBS programs and SCBUs/NICUs to develop and provide screening, testing, and follow-up protocols. The goal is to complete NBS for every newborn cared for in an SCBU/NICU at the earliest time in accordance with disease-specific screening windows, with the highest degree of reliability and using the fewest number of specimens.

## Overview of Changes

This guideline replaces the previous edition of the approved guideline, NBS03-A, published in 2009. Several changes were made in this edition, including:

- Condensed and combined tables to show the most relevant aspects for screening preterm, LBW, and sick newborns admitted to the SCBU/NICU
- Added discussion of screening diseases organized by type (eg, endocrinopathies) focused specifically on implications of NBS for preterm, LBW, and sick newborns
- Added new information covering the implications for this population of screening for diseases recently implemented in some NBS programs, such as SCID, LSDs, and point-of-care screening for CCHD
- Added a description of alternative protocols and suggested elements to evaluate when different collection protocols and testing paradigms are considered, in addition to the recommended serial-screening protocol and algorithm

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

## Key Words

Congenital anomalies, low birth weight, neonatal intensive care unit, newborn dried blood spot screening, newborn hearing screening, point-of-care screening, preterm birth, sick infants, special care baby unit

# Newborn Screening for Preterm, Low Birth Weight, and Sick Newborns

## Chapter 1: Introduction

This chapter includes:

- Guideline’s scope and applicable exclusions
- Background information pertinent to the guideline’s content
- Standard precautions information
- “Note on Terminology” that highlights particular use and/or variation in use of terms and/or definitions
- Terms and definitions used in the guideline
- Abbreviations and acronyms used in the guideline

The goal of newborn screening (NBS) is to detect newborns at increased risk for having a particular disease. Because screening is by definition not diagnostic, confirmatory testing is needed after a screen-positive result. Additionally, screening may provide false-positive and/or false-negative results, which NBS programs strive to minimize as much as possible. One focus area in this effort is screening of preterm, low birth weight (LBW), and sick newborns.

The recommendations made in this guideline emphasize:

- Using the newborn’s limited blood resources efficiently
- Collecting sufficient specimen (but no more than necessary) only as needed to minimize the stress for these medically fragile newborns
- Collecting specimens at the proper time to identify, refer, and treat affected newborns

However, the recommendations made in this guideline cannot predict or account for all scenarios that may arise before, during, and after admission to special care baby units (SCBUs) and/or neonatal intensive care units (NICUs). There are times when following admission screening recommendations for newborns in the SCBU and/or NICU may be impossible or ill advised. Clinical judgment should prevail.

### 1.1 Scope

This guideline covers best practices for personnel in the SCBU/NICU and the NBS laboratory, primary health care providers (HCPs), and NBS program follow-up personnel, in order to provide all preterm, LBW, and sick newborns with valid newborn dried blood spot (DBS) screening and point-of-care (POC) screening. Best practices are established with consideration of special circumstances that may affect the recommended specimen collection timing and/or screening test result(s) interpretations. Special circumstances include the newborn’s condition and treatments administered, maternal conditions and therapies, and other factors affecting the optimal screening windows for the specific screened diseases in this population. This guideline focuses on treatments, practices, and newborn conditions seen in the SCBU/NICU that are either known or suspected to interfere with NBS test results, consequences of the interference, and recommendations for avoiding or counteracting these problems.



This guideline is intended to provide guidance for ensuring rapid, consistent, and complete DBS and POC screening, including appropriate follow-up, to ensure early diagnosis and treatment for preterm, LBW, or sick newborns affected with a screened disease. If implemented, this guideline will assist NBS laboratories, NBS programs, and HCPs in:

- Minimizing the risk of a missed or delayed diagnosis and treatment for all screened diseases
- Minimizing the number of false-positive and/or false-negative NBS results in this population
- Optimizing the timing and minimizing the number of DBS specimen collections
- Optimizing the timing of and process for POC screening
- Defining essential elements of QA relevant to this guideline
- Providing education on the effects of SCBU and NICU treatments on newborn DBS screening
- Supporting clinicians in optimizing use of NBS program functions for timely interventions
- Identifying areas that need additional research

This guideline is not intended to provide general information on screening for all diseases (only information pertinent to the SCBU/NICU population) or dictate SCBU/NICU care practices. This guideline does not include:

- Discussion of data systems in SCBUs/NICUs that might facilitate implementing this guideline
- Details of individual diseases screened, beyond salient points unique to the preterm, LBW, and sick newborn population
- Recommendations about testing for X-linked adrenoleukodystrophy,<sup>5</sup> because there is not yet sufficient experience with screening newborns, particularly the subpopulation of preterm, LBW, and sick newborns

## 1.2 Background

NBS has expanded in recent years, with some programs now screening for more than 50 separate diseases. In 2006, a core panel of 29 diseases and 25 secondary diseases was recommended in the United States.<sup>6</sup> At the time of this publication, the US Recommended Uniform Screening Panel has been expanded to 34 core diseases and 26 secondary diseases, with more diseases being reviewed for addition to the newborn DBS screening panel.<sup>5</sup> Although screening programs vary around the world, many have mechanisms in place to decide whether a new disease should be added to their screening panel.<sup>7</sup> The terminology recommended in 2006 for core and secondary diseases is used in this guideline.<sup>6</sup>

Each disease on newborn DBS screening panels has its own optimal screening window, which is the time period between when the NBS marker analyte concentration indicates disease and when symptoms develop in the baby. The goal of NBS is to identify and treat every affected baby before symptom onset and before irreversible mental and/or physical damage result. Of approximately 4 million newborns screened each year in the United States, it has been estimated that more than 6500 babies are diagnosed with a disease detected through newborn DBS screening,<sup>8</sup> more than 5000 babies are diagnosed with hearing loss following newborn hearing screening,<sup>9</sup> and an additional 875 newborns with otherwise undiagnosed critical congenital heart disease (CCHD) are identified by pulse oximetry screening.<sup>10</sup> However, because of inconclusive and false-positive results, depending on the NBS program, disease, and testing methodology, as many as 10% or more of babies screened may need some degree of follow-up. Many of these babies are cared for in SCBUs/NICUs.<sup>11,12</sup>

There have been great advances in the care and treatment of preterm, LBW, and sick newborns and vast improvements to NBS programs, but more information is needed about the effect of newborn conditions and treatments on the best screening window for screened diseases. The effects of preterm birth and

## Chapter 3: Preanalytical Considerations for Dried Blood Spot Screening of Preterm, Low Birth Weight, and Sick Newborns

This chapter includes:

- Specimen collection issues
- Recommendations for DBS specimen collection timing, including SCBU/NICU algorithm
- Alternative recommendations for DBS specimen collection timing
- Special considerations for DBS specimen collection timing

### 3.1 Specimen Collection Issues

#### 3.1.1 Birth Weight and Blood Volume

Preterm newborns have blood volumes equal to about 80 to 90 mL/kg of their birth weight, a volume of about 45 mL in a 500-g extremely LBW newborn, which is readily depleted, because most extremely LBW newborns need multiple blood tests for various reasons in addition to NBS. The total volume of blood collected for newborn DBS screening generally ranges from 200 to 500  $\mu$ L. In the United States and the United Kingdom, the average DBS needs 50 to 100  $\mu$ L of blood, and most screening programs require four to five blood spots from each newborn.<sup>26</sup> If smaller blood spots are collected per local policy, more than five blood spots may be required to obtain the minimum amount of specimen needed for NBS tests (eg, 10 blood spots would be needed for smaller spots with 20  $\mu$ L of blood for a total of 200  $\mu$ L of blood collected). Although this amount of blood is not excessive, screening programs should be aware of limited blood resources in this population and request the least amount of blood needed to complete screening or follow-up testing for small preterm babies.

In addition, specimen collection by multiple heel sticks may be contraindicated for some preterm newborns. Although heel stick with direct application is the preferred source and specimen collection method, other sources of blood might be appropriate (see CLSI document NBS01<sup>27</sup>). In limited situations, umbilical cord blood, venous blood, and arterial blood might be appropriate for obtaining blood for newborn DBS screening.<sup>28,29</sup> However, umbilical cord blood specimens may not be suitable for all analyses.<sup>30</sup> Umbilical catheters may be used to obtain blood for newborn DBS screening, provided that the line is cleared of intravenous (IV) fluids, heparin, antibiotics, and other extraneous substances (see CLSI document NBS01<sup>27</sup> for additional information on unacceptable blood collection sites). See Additional Resource for additional information on specimen sources.

#### 3.1.2 Prescreening Treatments

##### 3.1.2.1 Transfusion

RBC transfusion is frequently performed in LBW newborns and babies admitted to SCBUs/NICUs. Although extracorporeal life support (ECLS) usually includes donor blood, it is not consistently identified as a transfusion for the screening laboratory. ECLS should be indicated as a transfusion by facilities when they submit DBS specimens. NBS programs should consider a DBS specimen collected after ECLS to be a post-transfusion specimen, so that newborns who have undergone ECLS will be followed up with the same protocols as other newborns who have undergone transfusion.

## Chapter 5: Postanalytical Activities for Newborn Screening Test Results Follow-up

This chapter includes:

- NBS program follow-up of positive NBS test results
- Roles and responsibilities for NBS follow-up after admission to the SCBU/NICU

### 5.1 Newborn Screening Program Follow-up of Positive Newborn Screening Test Results

Follow-up activities for NBS include initiating contact with the HCP or parent to communicate results and recommendations and providing active surveillance until NBS test results are resolved or the newborn is under treatment and management (see CLSI document NBS02<sup>94</sup>). There are multiple follow-up systems used by NBS programs to accomplish these tasks, many of which are centralized within the program. Specific roles and responsibilities vary by jurisdiction according to local legislation and certification processes. However, regardless of the system used, the responsibility for follow-up is shared among the HCP, designated NBS follow-up program coordinators, family, and medical specialists appropriate for the suspected disorder.

### 5.2 Roles and Responsibilities for Newborn Screening Follow-up After Admission to Special Care Baby Unit and/or Neonatal Intensive Care Unit

NBS for preterm, LBW, and sick newborns provides special challenges and responsibilities for physicians or HCPs caring for the newborns and for the follow-up personnel working within or on behalf of NBS programs. The following subchapters describe the roles and responsibilities that are defined for:

- Ensuring efficient NBS and follow-up
- Preventing babies from being lost to follow-up
- Maximizing the health benefit to the baby

#### 5.2.1 Physicians or Health Care Professionals in the Special Care Baby Unit and/or Neonatal Intensive Care Unit

Responsibilities of physicians or HCPs in the SCBU/NICU include:

- Providing newborn DBS and POC screening information to parents per local rules and regulations
- Maintaining up-to-date and accurate patient records of preanalytical NBS activities
  - Ensuring demographic information submitted with the newborn DBS specimen is complete and correct
  - Ensuring other specimen collection information submitted with the newborn DBS specimen is complete and correct
  - Alerting the NBS laboratory to clinical concerns or interventions that interfere with NBS tests, eg:
    - Meconium ileus
    - PN
    - Steroid administration

**NOTE:** To facilitate this communication, some NBS programs provide data fields in which to record this information being submitted with the newborn DBS specimen.

SAMPLE



CLINICAL AND  
LABORATORY  
STANDARDS  
INSTITUTE®

950 West Valley Road, Suite 2500, Wayne, PA 19087 USA

P: +1.610.688.0100 Toll Free (US): 877.447.1888 F: +1.610.688.0700

E: [customerservice@clsi.org](mailto:customerservice@clsi.org) [www.clsi.org](http://www.clsi.org)

PRINT ISBN 978-1-68440-052-2

ELECTRONIC ISBN 978-1-68440-053-9