

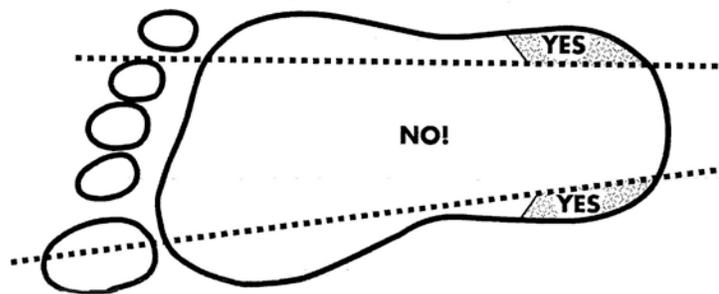
# TECHNIQUES FOR BLOOD COLLECTION ON FILTER PAPER

Collection of dried blood spots on filter paper should be only by the direct heel stick procedure. Reference the National Committee on Clinical Laboratory Standards (NNCLS), now known as Clinical Laboratory Standards Institute (CLSI): LA4-A5 "Blood Collection on Filter Paper for Newborn Screening Programs; Approved Standard - Fifth Edition. To obtain copies of LA4-A5 contact CLSI at [www.clsi.org](http://www.clsi.org) or e-mail [CustomerService@clsi.org](mailto:CustomerService@clsi.org) or phone 610-688-0100.

The following instructions are consistent with the recommendations in the Clinical and Laboratories Institute (CLSI) *Blood Collection on Filter Paper for Newborn Screening Programs; Approved Standard - 5<sup>th</sup> Edition* (CLSI LA4-A5).

## Sampling Technique

Collect the blood onto the labeled filter paper, using the following protocol:



1. Cleanse infant's heel with 70% isopropyl alcohol (use only rubbing alcohol).  
*Note:* Warming the skin-puncture site with a warm moist cloth, or a heel warming device, for 3 minutes can increase blood flow through the site.
2. **Allow heel to air dry.**
3. Using a lancet, or heel incision device, and wearing gloves, perform the puncture on the plantar surface of the heel (as indicated in the drawing). The puncture should be made to a depth of less than 2.0 mm with a sterile lancet or incision device.
4. Gently wipe off first drop of blood with sterile gauze or cotton ball. The initial drop contains tissue fluids that may dilute sample.

5. **Wait for formation of large blood droplet; apply gentle pressure with thumb and ease intermittently as drops of blood form.**
6. Gently touch the printed side of the filter paper card to the blood drop and in one step, allow a sufficient quantity of blood to soak through and completely fill a pre-printed circle. **Do not** press the filter paper against the puncture site on the heel. Fill each printed circle with a SINGLE application of blood. Observe both sides of the filter paper card to assure that blood uniformly penetrated and saturated the card. Spotting should be done *only* on the printed side. The filter paper must not touch the skin puncture site.
7. Fill the required number of blood spots for mandated tests.
8. All used items should be disposed of in an appropriate biohazard container.
9. Elevate infant's foot above the body and apply pressure using sterile gauze. Do not apply adhesive bandages.
10. Allow blood specimen to AIR DRY THOROUGHLY, on a horizontally level—non-absorbent open surface, such as a plastic-coated test tube rack—**for a minimum of 3 hours** at ambient temperature and away from direct sunlight. Do not stack, heat, or allow to touch other surfaces during the drying process.
11. **Within 24 hours of collection**, prepare the dried blood collection card for shipment to the laboratory by courier or mail. Only use the mailing or courier envelope provided. Do not use plastic or plasticene envelopes. Humidity and moisture are detrimental to stability of dried blood spot specimens and can affect results.

**Collection of NBS Specimen by Other Methods: Collection by Capillary Tube, Umbilical Arterial Catheter (UAC)**

Mechanisms to communicate to the designated screening laboratory specimen collection by other methods are not in place. Cut-offs at the laboratory are based on assumption that the specimen was collected via direct heel stick procedure. Cutoffs have not been established for specimens collected by alternate methods such as capillary tube, dorsal hand vein or umbilical catheter.

## MOST COMMON ERRORS IN SPECIMEN COLLECTION

INVALID SPECIMEN	POSSIBLE CAUSES
Quantity of blood not sufficient for testing (QNS)	Filter paper circles incompletely filled or not saturated/not all circles filled. Blood applied with needle or capillary tube. Contamination of surface of filter paper circle before or after specimen collection by gloved or ungloved hands, or by substance such as hand lotion or powder, etc.
Blood spots appear scratched or abraded	Blood applied improperly using capillary tube or other means (blotter has been damaged or torn by device).
Blood spots wet	Specimen not properly dried before mailing.
Blood spots appear supersaturated	Excess blood applied (usually with capillary tube or needle). Blood applied to both sides of filter paper.
Blood spots appear diluted, discolored, or contaminated	Puncture site squeezed or "milked." Exposure of blood spots to direct heat. Contamination of filter paper before or after specimen collection by gloved or ungloved hands, or by substances such as alcohol, formula, water, powder, antiseptic solutions, or hand lotion. Contamination during transit.
Blood spots exhibit "serum rings"	Alcohol not wiped off puncture site before skin puncture is made. Filter paper has come into contact with alcohol, water, hand lotion, etc. Puncture site squeezed excessively. Specimen dried improperly. Blood applied to the filter paper with a capillary tube.
Blood spots appear clotted or layered	Same filter paper circle touched to a blood drop several times. Circle filled from both sides of the filter paper.
Blood will not elute from the blotter paper	Blood specimen has been heat-fixed. Blood specimen is too old (more than two weeks between collection and receipt by the screening laboratory).