Guide for Receiving and Storing Dried Blood Spots (DBS) for Molecular Testing at Reference Laboratories

Initial Receiving and Processing Steps:



Upon arrival, examine the DBS package, test request form, and transport log. Ensure DBS cards meet the minimum standards for molecular testing:

- Identifying information on the DBS card is clear and matches the accompanying paperwork
- At least 3 circles are filled on the card
- All blood spots have a dark, uniform color
- DBS cards are properly packaged

Note: DBS cards for Early Infant Diagnosis (EID) and Viral Load (VL) or Drug Resistance (DR) testing should not be packaged in the same bag.



Assign and affix a unique laboratory ID number to each DBS card. Be sure to wear gloves, and do not touch the blood spots on the card.

Assign and affix the same laboratory ID number as the DBS card to each corresponding test request form.

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- The term Early Infant Diagnosis (EID) here is used broadly to include all virologic testing for the purpose of HIV diagnosis in infants and children <18 months of age.
- When receiving DBS for EID, VL and DR testing in the laboratory, handle DBS for EID in a separate area from DBS for VL and DR testing to avoid potential cross-contamination, as DBS for VL and DR testing are known HIV-positive specimens.
- It is recommended to change gloves frequently, especially when handling DBS from different sources.



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Storage Steps:

These procedures may be used for:

- Storage of the remaining DBS after testing and documentation have been completed
- Storage of DBS which may not be tested within 2 weeks of specimen collection

Notes

- DBS should be stored separately according to the type of testing to be performed: diagnostic (EID) or monitoring (VL or DR).
- Maintain EID DBS and VL/DR DBS in separate bags and separate freezer boxes; when possible, also use separate locations within the freezer(s).



Wrap each DBS card in glassine paper or insert into a glassine envelope.



Package the DBS cards in zip closure plastic bags.

- Separate the wrapped EID, VL and DR DBS cards and place up to 10 individually wrapped EID, VL, or DR cards in a low gas-permeable, zip closure plastic bag.
- Insert 10 small desiccant packs and a humidity indicator card in each plastic bag.
- Expel any air within the bag and seal tightly. Double check that the closure is secure.









Clearly label the outside of each bag with the date, range of specimen numbers, and type of test (EID, VL, or DR). If EID specimens have already been tested, note whether they are HIVpositive or negative samples.



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Place 10 bags in a freezer box (13X13cm). On each box, write the box number, the range of lab ID numbers, and number of bags. Transfer this information to the Lab Data Log.



Notes

- Remember to store EID samples separately from VL and DR samples.
- To prevent cross-contamination among EID specimens that have already been tested, it is also recommended to store EID positive DBS cards separately from EID negative DBS cards.



Place the boxes in a -70° C freezer dedicated for sample storage (no PCR products and reagents).



Monitor DBS Storage conditions: For long-term DBS storage, monitor the stored DBS packages for humidity changes daily during the first week of storage and monthly thereafter. Minimize freeze-thaw cycles (up to 3 freeze-thaw cycles are acceptable).



- Consult the humidity indicator card manufacturer's instructions to determine criteria to detect humidity exposure (e.g. color change).
- If DBS cards have been exposed to humidity, inspect the bag for holes and proper sealing, and replace bag if necessary, or change desiccants and humidity indicator card.
- If the humidity indicator card signals humidity exposure to
 - □ 30%: this is a warning that the DBS card has been exposed to humidity and should be monitored closely
 - 40%: the humidity indicator card and desiccant bags should be replaced with new ones
 - □ 50%: the DBS samples should be examined for quality and humidity exposure
- It is imperative to record the storage conditions found after each inspection.
- Refer to national or local guidelines on the maximum length of storage of DBS samples.
- Once results are approved and released, specimens may be removed for destruction.



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Dried Blood Spot (DBS) Laboratory Acceptance Criteria for Polymerase Chain Reaction (PCR) Testing

This job aid is a guide for laboratorians handling DBS in PCR testing laboratories

Each DBS sample tested must meet the following criteria:



2.

Identifying information on the DBS card must be clear and match accompanying paperwork.

At least 3 FULL CIRCLES must be obtained. A circle is full once the blood has just reached the dotted line or perforation.



DBS should be dark and uniform in color, indicating proper collection and drying technique.

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