Nested serosurvey study

Standard Operating Procedures

Specimen Collection: Dried Blood Spot

Version: November 6, 2016

1. Purpose/Background

1.1. The purpose of this document is to describe how to collect a blood sample from a finger prick in the form of liquid blood or dried blood spots (DBS).

2. Scope/Applicability

2.1. This SOP is intended for all individuals who will collect blood as part of study procedures.

3. Roles and responsibilities

3.1. Data collectors for the serosurvey are responsible for blood collection.

4. Prerequisites / Supplies Needed

4.1. Ensure you have all the material that you need BEFORE beginning:

- Gloves
- Methylated spirits / alcohol
- Cotton swabs
- Blue adult lancets (BD Microtainer Contact-Activated Lancet. 2 mm depth)
- Pink child lancets (BD Microtainer Contact-Activated Lancet. 1.8 mm depth)
- For dried blood spot collection:
    - Dried blood spot (DBS) cards
    - DBS drying rack
    - Ziploc storage bags
    - Dessicant packs
- For liquid capillary blood collection:
    - Microtainer tubes
    - Tube rack/holder
    - Cold box
    - Freezer packs
- Sharpie pen
- Plastic container to transport the DBS card
- Sharps container
- Biohazard bag
- Table and chair, optional

5. Procedural steps

- Arrange blood collection materials (alcohol, cotton swabs, lancet, microtainer tube, DBS card) in a clean area. Place biohazard bag, sharps container, cold box, and DBS drying rack nearby.

- Using the pen, label 1 DBS card, 1 ziploc bag, and 1 microtainer tube with the date (DD/MM/YY) and participant's study ID: Cluster (2 digits) – HH (2 digits) – Individual (2 digits)
- Remove the cap of microtainer tube and place the cap aside in a clean location.
- Put on a pair of gloves

For blood collection with children under age 8:

6a.1. Ask the caregiver to sit on a chair with the child on his/her lap and to warm the child's hand by holding and rubbing it.

6a.2. Show caregiver how to immobilize the child:

- Position guardian's legs around the child's legs in a cross-leg pattern
- Ask the guardian to extend their arm across the child's chest and tuck the child's free arm underneath. With that same hand, the guardian should grasp the child's other arm at the elbow (i.e. the hand to be pricked), and hold it securely
- Ask the guardian to use their other arm to firmly grasp the child's wrist, holding it palm down so that the fingers are below the elbow, tilted towards downwards.

For blood collection with children age 8 and older:

6b.1. Ask the participant to sit on a chair with his/her hand extended on the lap so that fingers are below the elbow and palm is face upward.

1. Pricking the finger (video tutorial link)
   - Explain the procedure: *"I am now going to prick your/your child's finger to collect the blood in this tube and on these papers. This may hurt temporarily but it will be quick and will not cause any harm. Do you have any questions before we begin?"*
   - Select a finger without abrasions and few callouses. The 4th finger (ring finger) is the top choice. If not, select the 3rd finger (middle finger). Do NOT attempt on the 1st finger (thumb) or the 5th finger (pinky).

   - Wash gloved hands with methylated spirit /alcohol.
   - Wipe the area of selected finger with spirit/alcohol swab and allow to air dry.
   - Select the appropriate lancet (pink for age < 8 years old, blue for age 8 and older)

   - Twist the cap of the lancet until it is removed
   - Place the lancet on the side of the ball of the finger perpendicular to the line of the fingerprint.
   - Hold the finger firmly close to the planned puncture location. Press firmly on the finger when you press the button and lance the finger (across the fingerprint) between the side and the pad. Doing so will help you to obtain the amount of blood you need.
   - Gently rub the hand or finger to maintain blood flow. **Do NOT tightly squeeze the finger.**
   - Wipe the first drop of blood with a cotton swab

- **Collecting blood spots on DBS cards**
    - Place DBS card under the finger and **let the blood droplets form and drop onto the first circle**. Do not wipe the DBS card on the finger.
    - **Do NOT start next circle until the first circle is completely filled** (no white visible inside circle)**.** When 1st circle is completely full, drop blood onto next circle. Continue until 5 circles are completely filled. See Appendix 2.
    - If liquid blood is also being collected, continue to hold the finger and maintain blood flow. If not, apply firm pressure with a sterile cotton swab to stop the bleeding.
- **Collecting liquid blood in microtainer tube**
    - Place capillary blood tube horizontally or at a slight incline.
    - Let blood droplets form and fall into the tube. **Do NOT let the blood flow across the finger. Do NOT scrape the tube against the finger to collect blood.**

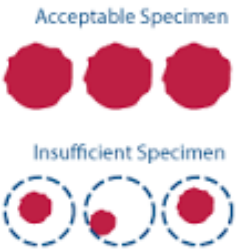
1. Fill tube to 0.25 mL (250 uL). Do not exceed more than 0.5 mL (500 uL). Put top on tube and put in tube rack.
2. When finished, apply firm pressure with a sterile cotton swab on the finger to stop the bleeding.

- **Pricking finger more than once.** The data collector may make up to 3 fingerprick attempts if:
    - Insufficient blood is collected. See Troubleshooting tips to increase blood flow (below).
    - The data collector misses puncturing the finger. If this occurs, ensure the caregiver is holding the child's arm firmly against a flat surface to steady the puncture site.
    - Interviewer can attempt finger prick **maximum three times**. For each attempt, select a new different finger.

- **Clean up and storage**
    - Place DBS card on drying rack. Let dry for 20-30 minutes and then place in a Ziploc bag.
    - Double check the cap is secure. Mix the specimen by gently inverting tube upside down 3 times.
    - Confirm participant ID on tube is the same as participant ID on household census.
    3. Place labeled tube in cold box and replace lid.
    - Dispose of lancet in sharps container. Dispose gloves, cotton balls, and spirit in biohazard bag.
- **Transport and long-term storage**

4. Transport the DBS cards and microtainer tubes in the cold box to the field supervisor at the end of the day.
5. DBS cards need to dry on a drying rack overnight in a dust-free area. Return DBS cards to the same Ziploc bag with 1 dessicant pack. Samples will be shipped every 1-3 days to Macha Research Trust for storage in -20 ^{o}C untl testing.
6. Microtainer tubes of blood need to be transferred to Macha Research Trust within 24 hours for processing and storage.

Appendix 1. Appropriate blood collection on a DBS card



TROUBLESHOOTING

Potential Roadblocks for Blood Sampling	**Proposed Action(s)**
What if a surveyor runs out of materials?	Contact the field supervisor. If the field supervisor runs out of materials, contact the study coordinator.
What if the cold packs are no longer cool?	Contact the field supervisor. Cold packs should be used for a maximum of 24 hours.
What if the child is agitated, crying, or will not sit still?	For children, ask the caretaker or another member of the household to hold the child (see #6 above). Reassure the child or distract the child with a toy or video.
What if the guardian requests that we stop the sampling?	The participant or parent/caretaker have the right to stop blood collection at any time. If requested, stop the procedure and apply firm pressure with a sterile cotton swab to stop the bleeding.
What if the lancet does not prick the finger or is otherwise broken?	Dispose of it into the sharps bag. Use a new lancet to attempt pricking the child's finger again.
What if the blood collects at the top of the microtainer tube?	Tap the tube on the table to draw the blood down into the tube and avoid coagulation. If this does not work and there is not sufficient enough blood, dispose of tube in a biohazard container. Using a new lancet, the data collector should select a new finger to attempt blood collection again (maximum 3 attempts)
What if the surveyor cannot collect enough blood?	The surveyor may make 3 attempt at the fingerprick using a fresh lancet each time and a different finger. Prior to the fingerprick: 1. Have caregiver hold the child's hand below the heart, let it hang, and then shake it gently 2. Rhythmically tighten and release the child's wrist, to ensure that there is sufficient flow of blood. 3. Keep the child warm by removing as few clothes as possible, swaddling an infant in a blanket, and having the caregiver wrap around the child for warmth, leaving only the extremity of the site of capillary sampling exposed. During blood collection: 1. Gently rub the hand or finger to maintain blood flow. Gently rub the hand or finger to maintain blood flow. Do NOT tightly squeeze the

	finger. After 3 attempts, do not attempt again.
What if the data collector injures him/ herself with the lancet?	1. Follow the Post-Exposure Prophylaxis protocol 2. Report the incident to the Field Coordinator immediately
What if the data collector's nose, mouth or skin (broken or not) make contact with the blood sample?	3. Follow the Post-Exposure Prophylaxis protocol 4. Report the incident to the Field Coordinator immediately
What if the participant loses consciousness, begins to sweat, becomes pale, vomits, sighs or stares excessively, convulses, or has a sudden drop in blood pressure during or after having their finger pricked?	In the rare circumstance that the participant loses consciousness, begins to sweat, becomes pales, vomits, sighs or stares excessively, or has a sudden drop in blood pressure: 1. Discontinue the blood sampling immediately 2. Lay the participant down and place them in a recovery position (i.e. head to the side and chin up). 3. Elevate their feet above their heart (~30 cm) 4. Check that the child's airway is clear by first visually inspecting the child for the rise and fall of their chest. Then, you may place your cheek near the child's mouth and nose to feel their breath. 5. Loosen any belts, collars, or other tight fitting clothes 6. If the child vomits while unconscious, clear the airway with finger and check again for breathing. If participant stops breathing: 1. Instruct someone to call for a physician 2. Perform CPR until a physician or other medical aid arrives If the participant does not regain consciousness after one minute or begins to convulse: 1. Call a physician or provide transport to a medical facility 2. Prevent the participant from standing because they are at risk of fainting again. If the participant regains consciousness quickly: 1. Monitor the participant for 10 minutes after the event 2. Give fluids to drink AFTER the child has received the proper care: 3. Note the event on the household census 4. Thank the family for their time