I. Purpose of Procedure (Principle)

To standardize the process for obtaining the maximum volume (up to 20 milliliters) of blood from a patient for Blood culture testing while observing aseptic technique.

II. Scope

Dartmouth-Hitchcock Laboratory staff that collect blood culture samples

III. Definitions

Unique patient demographic identifiers: Name, Date of Birth (DOB), medical record number (MRN), Social security number (SS#)

Adult blood culture set: Consists of (1) aerobic (Blue bottle) and (1) anaerobic (Lavender bottle) each bottle holds up to 10 mL of blood

Pediatric blood culture: Consists of (1) (Pink bottle) holding up to 4 mL of blood.

Isolator blood cultures: Fungal isolator blood cultures used for the detection of dimorphic fungi (primarily Histoplasma)

MB BacT blood culture bottle (black bottle): Used to culture Acid Fast Bacilli (AFB); Mycobacteria

FAN: Fastidious Antibiotic Neutralizations

IV. Specimen Requirements

A. Specimens are required to have at least two unique identifiers, with the name being one of them (unique identifiers are indicated below with an asterisk)

- Patient name*
- Patient MRN*
- Patient date of birth*
- Collection date

B. Draw blood cultures prior to the start/change of antibiotics

1. In emergency situations where the administration of antibiotics cannot wait, cultures are drawn soon as possible.
2. If blood cultures are required during a course of antibiotics, blood cultures should be drawn when the antibiotics are at their lowest levels.

V. Equipment, Reagents and Required Records/Forms

A. Personal Protective Equipment

All employees must adhere to Universal precautions: Treat all body specimens as if they are potentially infectious. Use Personal protective equipment. Refer to the policies for Personal Protective Equipment (PPE) on the D-H Health and Safety Manual in the procedural assistance.

Wear lab coats and gloves whenever potentially infectious materials may be generated or contamination is reasonably anticipated. Use eye protection devices such as goggles or glasses with solid side shields, or chin- length face shields whenever splashes, spray, spatter or droplets of blood or other potentially infectious materials may be generated and eye, nose or mouth contamination can be reasonably anticipated.

B. Equipment

- Adult BacT/Alert blood culture set or one Pedi bottle.
- Cleansing solution:
  - Adults: ChloraPrep One-Step Frepp Applicator
  - Infant less than 2 months of age: 70% propyl-alcohol pads
  - Infant less than 2 months of age: Povidone-iodine swabs
- Sterile 3 mL or 5 mL syringe
- 1 butterfly needle.
- Luer adapter, Lg. culture bottle vacutainer holder.
- Blood culture transfer device
- Gauze
- Tourniquet
- Paper tape

C. Reagents – N/A

D. Required Records/Forms

Laboratory Requisitions completed with patient demographics, test orders and diagnosis codes and ordering provider.

VI. Equipment Calibration/Process Validation

System validation on file in the Laboratory Information Systems (LIS) office

VII. Quality Control/Process Control

Ensure that labels are affixed properly and match patient information

VIII. Procedure and Calculations

A. Bottle measurements and Volume of Blood: Volume is critical to most accurately detect bacteremia, especially if patient is already receiving antimicrobial therapy. In infants and children, the concentration of organisms during bacteremia is higher than in adults, so less blood is required for culture.

Note: For low volume (less than 10 mL) collected on a adults, **do not** split the amount between the aerobic and the anaerobic bottles instead use only the adult Aerobic (Blue) bottle

1. **Adult Blood Culture Bottles** are marked with 5 mL graduations on the side of the bottles.
   a. Use the lines to estimate the amount of blood entering the bottle.
      - Total volume for adults is 16- 20 mL of blood per venipuncture divided evenly into aerobic and anaerobic bottles
      - Fill to the 10 mL mark using the bottle measurement indicators
      - Do not overfill

   Note: The vacuum of the bottles is strong enough to overfill the container.

2. **Pediatric Blood Culture Bottles** are for patients under the age of 11. Pediatric bottles drawn on adults are accepted, but not ideal.
   a. Bottle measurement lines are in increments of 4 mL
      - Total volume for children and infants: 0.5 to 5 mL of blood per venipuncture
      - Fill with up to first line of the 4ml bottle measurement indicators
      - Do not overfill

B. Procedure:

1. Verify provider order for blood culture collection and patient identity.
2. Assemble supplies appropriate to patient's size. Use the largest bore butterfly possible to increase blood flow.
3. Apply tourniquet to extremity and palpate vein with clean, gloved hands.
4. Remove tourniquet.
5. Prep patient skin with appropriate skin cleansing solution.
   a. Adults: Open the ChloraPrep One-Step Frepp kit
      i. Squeeze tabs together to break open ampule
      ii. cleanse the area in outward circular motion with the sponge for 30 seconds
      iii. allow to air dry
   b. Infants less than 2 months age:
      i. Vigorously cleanse the site with 1 alcohol pad for 30 seconds and allow to air dry
      ii. Once alcohol has dried, cleanse the site with 3 povidone-iodine swabs:
         - Apply swabs in concentric circles
         - allow to air dry

6. Remove plastic top from blood culture collection bottles (aerobic, anaerobic, or pediatric)
7. Wipe rubber diaphragm with isopropyl alcohol swab, remove and let diaphragm air dry. Keep the bottle tops away from other surfaces to eliminate contamination.
8. Apply tourniquet to extremity without compromising site
9. Using appropriately sized butterfly needle and syringe, puncture vein and withdraw volume of blood appropriate for patient age and size (pediatric 0.5 -5mL, adults 16 - 20 mL)

    a. **Vacutainer Collection Method**
        i. Attach a vacutainer tube holster to the butterfly. This may be used as a separate entity, or a blood culture adaptor may be attached to the butterfly setup.
        ii. Insert needle into patient's arm
        iii. While holding the vacutainer tube holster insert the aerobic blood culture bottle and allow the bottle to draw 10 mL of blood.
        iv. Remove the aerobic bottle and insert into anaerobic bottle. Allow to fill with 10 mL.
        v. If additional blood work is ordered, insert the appropriate vacutainer tube by order of draw.
        vi. Fill all remaining blood tubes.
    b. **Syringe collection Method**
        i. Attach a syringe to the butterfly collection set.
        ii. Insert needle into patient's arm
        iii. Draw off 20 mL of blood using syringes.
        iv. If additional blood work is required, you have an option of collecting by using additional syringes or performing an additional venipuncture.
        v. Attach a blood culture transfer device to the syringe and insert into the aerobic bottle. Allow 10 mL of blood to flow into the bottle.
        vi. Attach a new transfer device to the syringe and repeat the procedure using the anaerobic bottle. Discard the butterfly and syringe combo.

    Note: If venipuncture is unsuccessful, use a new supply setup for the reattempt

    Note: This procedure must be performed aseptically. Pay special attention to technique to maintain the sterility

    Note: Care should be taken to avoid air entering the anaerobic blood culture bottle, ensure residual air from the syringe is not pushed into the anaerobic bottle

    Note: Do not force sample into the bottle.

10. When inserting blood into anaerobic bottle, avoid air being injected into bottle
11. Place syringe and transfer device into sharps container.
12. Label specimen using provided labels on printed slip.
13. Fill in appropriate areas on requisition slip including:
    a. Date/time drawn

b. Location of site
c. Name or initials of person drawing the sample

14. Place bottles into biohazard bag with specimen label.

15. Send to microbiology using the pneumatic tube system.

IX. References

- Clinical and Laboratory Standards Institute (CLSI), Principles and Procedures for Blood Cultures: Approved Guideline, 2007. CLSI document M47-A (ISBN 1-56238-641-7)
- Baron, E., Weinstein, M. et al. Blood Cultures IV. Procedures in Clinical Microbiology, ASM, 2005.
- Garza, D., Becan-McBride, K. Phlebotomy Handbook Blood Specimen Collection from Basic to Advanced, 8th Edition, 2009, Prentice Hall.