Blood Culture – Skin Preparation and Specimen Collection

Purpose

This procedure is intended for preparing venipuncture site and blood culture bottles, and collecting blood specimen, using proper phlebotomy/venipuncture technique, when Blood Cultures are ordered by providers.

Scope

This procedure may be performed by the trained laboratory assistants, phlebotomists and other health care providers responsible for the collection and handling of blood specimen from inpatients and outpatients.

Policy

Timing of collection:

- Most cases of bacteremia are detected by obtaining three sets of separately collected blood cultures. More than three sets of blood cultures yield little additional information
- A set of blood cultures consist of an aerobic (SA/blue cap) and an anaerobic (SN/purple cap) blood culture bottle.
- Blood cultures should be drawn prior to the start of antibiotics.
- Blood cultures must be drawn at the time specified by the ordering provider.
- When more than one set is ordered at the same time, the second set should be obtained from a separate and prepared venipuncture site on the opposite arm, within 30 minutes after the first set is collected.
  Note:
  In medical and critical urgent situations in which antimicrobial therapy is to be started immediately, two or three separate blood culture specimens can be collected from different sites within a few minutes apart.
- There should be no more than three sets of blood cultures drawn within a 24 hour period.

Sample Collection:

- Laboratory policy allows two (2) attempts per phlebotomist. If the first and second attempts are unsuccessful, another phlebotomist must draw the patient. If still unsuccessful, the nurse in charge must be notified.
- If a patient has an intravenous (IV) on one arm, the phlebotomist must obtain the blood specimen from the other arm without an IV.
- Blood Culture specimen bottles must be labeled in the presence of the patient after every blood collection.
- ChloraPrep® must **ONLY** be used for patients greater than 60 days old to disinfect the venipuncture site.
- For newborn/infant (neonate) patients less than 60 days old, 70% isopropyl alcohol prep pad, sterile saline, or soap and water must be used to disinfect the venipuncture site.

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Specimen requirements

Adults:

- Each Blood Culture request consists of a minimum of 2 (two) sets of separately drawn blood specimens from two different venipuncture sites.
    - 1 set = 2 blood culture bottles, one aerobic (SA/blue) and one anaerobic (SN/purple)
- Total volume required: 8-10 mL for each blood culture bottle from each venipuncture site.
  Note: Adult adequate fill volume is crucial in the diagnosis of sepsis.

Pediatrics:

- Collect 1 to 2 sets of blood cultures from **1-2 different venipuncture sites**.
    - 1 set = 2 blood culture bottles, one aerobic (SA/blue) and one anaerobic (SN/purple)
- Volume required: 2-4 mL for each blood culture bottle per venipuncture site.
  Notes:
  Neonates/infants (less than two months old) – when less than 1 mL of blood is collected, make sure that the actual volume is written directly on the blood culture bottle, and recorded in the laboratory systems' Notes (not in Comments) so that the information would be available/viewable to the ordering provider in KPHC.
- When less than 1.0 mL of blood is collected, the priority is to inoculate the aerobic (SA/Blue) blood culture bottle.

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Blood Culture – Skin Preparation and Specimen Collection, Continued

Materials

- Disposable powder-free latex or latex free gloves
- Tourniquets – single-use disposable latex free
- Antiseptics for skin preparation
  - ChloraPrep® (Chlorhexidine gluconate) prep applicator (sterile sponge and ampule)
  - 70% isopropyl alcohol prep pads (also used to clean the tops of the blood culture bottles)
  - Sterile saline
  - Soap and water
- BacT/ALERT® blood culture media – plastic bottles
  - Aerobic (SA/blue cap)
  - Anaerobic (SN/purple cap)
- Winged (Butterfly) collection set – for direct draw
- Saf-T Holder Blood Culture Device with Male Luer Adaptor for use with winged collection set
- Sterile disposable syringes: 3, 5, 10, or 20 mL, with 21-23 gauge needles
- Saf-T Holder Blood Culture Device with Female Luer Adaptor for use with syringe
- Sharps container – OSHA acceptable puncture resistant disposable container, with lid marked clearly as a biohazard.
- Adhesive bandages or tape
- Gauze pads
- Blood collecting trays or carts

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Blood Culture – Skin Preparation and Specimen Collection, Continued

Before you begin

- Wash hands with soap and water, or an alcohol-based hand rub. Always wear properly fitting gloves.
- Use latex-free gloves and tourniquet if a patient states they have a latex allergy.
- Mark the 10 mL line on the blood culture bottles
  - Blood culture bottles have vacuum, but it is not measured as in evacuated tubes. Avoid overfilling by marking the fill level on side of bottles prior to collection. (Overfilled bottles may cause false positive readings.)
- Prevent or Avoid Contamination of blood culture samples when multiple tests are requested by first inoculating the blood culture bottles, and then the other required blood collection tubes.
- Observe Universal Precautions when obtaining sample.

Skin preparation for Patients older than 60 days

Follow the steps below to decontaminate the venipuncture site for patients older than 60 days using ChloraPrep® applicator.

Step	Action
1	Select the venipuncture site to be used to collect blood sample as follows: • Apply the tourniquet and locate the vein to be used. • Remove the tourniquet.
2	Open ChloraPrep® pouch and remove applicator. ***Note:*** *Applicator is for single use only. Discard after single use.*
3	Pinch the wings on the applicator to break the ampule and release the antiseptic. • **DO NOT** touch the sponge. • Wet the sponge by repeatedly pressing and releasing the sponge against the treatment area until liquid is visible on the skin.
4	Use the wet sponge to cleanse the venipuncture site, using back and forth strokes for 30 - 60 seconds, covering an area of 2.5 x 2.5 inches.
5	• Allow the decontaminated area to air dry completely (which may take from 30 - 60 seconds). • **DO NOT** blot, wipe away or blow dry.
6	Proceed to collect the specimen for Blood Cultures.

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Skin preparation for patients 60 days old or less

Follow the steps below to decontaminate the venipuncture site for patients 60 days old or less using 70% isopropyl alcohol prep pad, sterile saline or soap and water.

Step	Action
1	Use the 70% isopropyl alcohol prep pad (sterile saline or soap and water may be used in lieu of alcohol) to cleanse the venipuncture using back and forth strokes for 30 seconds
2	• Allow the site to air-dry. • **DO NOT** BLOT, WIPE OR BLOW DRY. ***Note:*** *Allowing the puncture site to air dry prevents specimen hemolysis and burning sensation to the patient when the needle is inserted.*

Blood collection using winged collection set

Follow the steps below for collecting blood samples and inoculating Blood Culture bottles using a winged collection set.

	Bottle Preparation
Step	**Action**
1	• Inspect the bottle surface, the media, bottom sensor and expiration date. • Ensure that the broth is clear and the sensor is intact and a blue-green color. • **DO NOT** USE bottle if the media exhibits turbidity, the sensor is yellow, or the flip top seal was removed. Discard the bottle.
2	• Remove the flip-top covering on the bottles. • **DO NOT** remove the flip-top until ready to collect blood.
3	• Clean the top of the blood culture bottles with alcohol pad. • Set the bottle upright after cleansing. ***Note:*** *Always use fresh alcohol pad to clean top of each bottle*.
4	Attach the winged collection set to the Male Adaptor.
5	Reapply the tourniquet and insert the needle into cleansed vein as described in the Blood Collection Venipuncture procedure. ***Note: DO NOT*** *re-palpate the vein/prepared skin area.*

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Blood collection using winged collection set, continued

<table>
<tr><th></th><th>Venipuncture and Bottle Inoculation</th></tr>
<tr><th>Step</th><th>Action</th></tr>
<tr><td>6</td><td>
• Fill the aerobic (SA/blue) bottle first.
  • Place the Blood Collection Adapter Cap on the bottle septum.
  • Press down to penetrate and obtain blood flow.
  • While holding the adapter cap, verify that blood flows into the bottle.
  • Verify that sufficient amount of blood collects into the bottle.
• Fill the anaerobic (SN/purple) bottle by following the same steps as the aerobic (SA/blue) bottle above.

Note: The recommended volume for adults is 8-10 mL of blood per blood culture bottle.

<table>
<tr><th>If drawing…</th><th>Then…</th></tr>
<tr><td>Adult</td><td>Collect 8 to 10 mL of blood for each blood culture bottle.</td></tr>
<tr><td>Pediatric samples, Neonates, and difficult venipuncture</td><td>• Collect a minimum of 1 mL of blood into the aerobic bottle (blue). Recommended fill is 2 to 4 mL.
• For difficult venipuncture, document on the label that it is a “hard stick”.

Note: Due to difficulty in specimen collection, for neonates and/or nursery samples it is acceptable to inoculate the aerobic (SA/blue) blood culture bottle with any amount of blood that is collected.</td></tr>
<tr><td>Additional blood for other tests</td><td>Collect the appropriate samples after collecting the blood sample for the Blood Culture bottles.

Note: Always collect or draw blood for Blood Culture bottles first.</td></tr>
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Blood collection using syringe

Follow the steps below for collecting blood samples and inoculating Blood Culture bottles using a syringe.

	Bottle Preparation
Step	**Action**
1	• Inspect the bottle surface, the media, bottom sensor and expiration date. • Ensure that the broth is clear and the sensor is intact and a blue-green color. • **DO NOT** use bottle if the media exhibits turbidity, the sensor is yellow, or the flip top seal was removed. Discard the bottle.
2	• Remove the flip-top covering on the bottles. • **DO NOT** remove the flip-top until ready to collect blood.
3	• Clean the top of the blood culture bottles with alcohol pad. • Set the bottle upright after cleansing. ***Note:*** *Always use fresh alcohol pad to clean the top of each bottle.*
4	Attach the safety needle to the syringe.
	Venipuncture and Bottle Inoculation
5	Reapply the tourniquet and insert the needle into cleansed vein, as described in the Blood Collection Venipuncture procedure. ***Note: DO NOT*** *re-palpate the vein/prepared skin area.*
6	Slowly pull back on the plunger to fill the syringe.
7	Release the tourniquet, remove the needle from the vein and activate the safety device.
8	Remove the needle from the syringe and discard in the Sharps container.
9	Attach the syringe to the Female Leur Adapter.

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Blood Culture – Skin Preparation and Specimen Collection, Continued

Blood collection using syringe, continued

	Venipuncture and Bottle Inoculation, continued
Step	**Action**

10	• Fill the anaerobic (SN/purple) bottle **first**.
	  • Place the Blood Collection Adapter Cap on the bottle septum.
	  • Press down to penetrate and obtain blood flow.
	  • While holding the adapter cap, verify that blood flows into the bottle.
	  • Verify that sufficient amount of blood collects into the bottle.
	• Fill the aerobic (SA/blue) bottle by following the same steps as the anaerobic (SN/purple) bottle above.
	
	Note: *The recommended volume for adults is 8-10 mL of blood per blood culture bottle.*

If drawing…	Then…
Adult	Collect 8 to 10 mL of blood for each blood culture bottle.
Pediatric samples, Neonates, and difficult venipuncture	• Collect a minimum of 1 mL of blood into the aerobic (SA/blue) bottle. Recommended fill is 2 to 4 mL. • For difficult venipuncture, document on the label that it is a “hard stick”. ***Note:*** *Due to difficulty in specimen collection for neonates and/or nursery samples, it is acceptable to inoculate the* ***aerobic (SA/blue) blood culture bottle*** *with any amount of blood that is collected.*
Additional blood for other tests	Collect the appropriate samples **after** collecting the blood sample for the Blood Culture bottles. ***Note: Always collect or draw blood for Blood Culture bottles first.***

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Sample handling - post collection

Follow the steps below to complete the blood collection.

	After Blood Collection is Complete
Step	**Action**
1	Gently invert blood culture bottles three times to prevent clotting.
2	Remove and dispose of the needle as described in *Blood Collection Venipuncture* procedure.
3	**Post Phlebotomy Puncture Site Care** After blood specimen collection is complete: • Apply pressure to the site for 5 minutes with a clean gauze pad. • Examine the patient's arm to determine if bleeding has stopped. • If so, apply adhesive or hypoallergenic tape over the gauze pad. • Instruct the patient to apply pressure on the puncture site for 5 minutes using their other hand. • Adhesive may be removed after 30 minutes.
4	Label the blood culture bottles with the pre-printed label that has the required patient identification information. • Patients full name • Patient's medical record number (MRN) • Date and collection time • Initials of person/NUID who performed the specimen collection procedure
5	• Place the accession label sideways on each blood culture bottle, making sure that the manufacturer barcode labels are not covered. • Make sure all specimens are entered in the specimen tracking system. ***WARNING:*** ***DO NOT cover the barcode labels on the blood culture bottles.***

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Specimen stability

- Inoculated blood culture bottles are stable at room temperature for 24 hours.
- Blood cultures received at the Regional Reference Laboratories greater than 24 hours after collection are not valid and will be rejected.
- After inoculation, blood culture bottles should **remain at room temperature** until transported on the next courier from the Medical Centers to the Regional Reference Laboratories.

Specimen transport

- Make sure that the specimens are accessioned and are entered in the specimen tracking system per protocol.
- Send the blood culture specimens immediately to the Regional Reference Laboratories with the next available courier.
- **DO NOT** incubate blood culture bottles prior to transport.

Controlled documents

The following controlled documents support this procedure:

Document Title
Performing Venipuncture
Standard Precautions and Safety Practices in the Laboratory
Infection Control
Universal Body Substance Precautions
Handling of Regular and Infectious Waste
Cleaning Work Areas
Proper Hand-Washing
Medical Center Media Plating Chart – Summary
Summary of Medical Center Media Plating Chart – With Temperature and Time Requirements
CERNER (ILIDS) Microbiology – Specimen Labeling Diagrams and Photos

Non-controlled documents

The following non-controlled documents support this procedure:

- Principles and Procedure for Blood Cultures; Approved Guideline, CLSI, May 2007
- Procedures for the Collection of Diagnostic Blood Specimens by Venipuncture, 6th Edition, CLSI, October 2007
- BioMerieux Blood Culture Collection with BacT/Alert Blood Culture System (Video/Package Insert)

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Reviewed and approved by (for Medical Center Area Approval Only):

SIGNATURE	DATE
Name: _____ **Operations Director, Area Laboratory**	
Name: _____ **CLIA Laboratory Director**	

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HISTORY PAGE

Type of Change: New Major, Minor	Description of Change(s)	Quality Systems Leader/Date	Operations Director, Area Laboratory Review/Date	CLIA Director Review/Date	Date Change Implemented
New					4/6/2016

Signature Manifest

Document Number: SCPMG-PPP-0114 **Revision:** 01

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All dates and times are in Pacific Standard Time.

Blood Culture – Skin Preparation an

Final Approval

Name/Signature	Title	Date	Meaning/Reason
Darryl Palmer-Toy (T188420)	SCPMG Laboratory Sys Med Dir	05 Apr 2016, 05:05:11 PM	Approved

Set Effective Date

Name/Signature	Title	Date	Meaning/Reason
Rebecca Rosser (K053260)	RRL ED CONSULTANT	06 Apr 2016, 08:56:40 AM	Approved