

PURPOSE: To determine the presence or absence of any bacterial organisms in transfusion products such as blood and platelets that may have become contaminated at the time of collection from donors, during processing or at the time of infusion into the patient. Any organism isolated must be considered significant.

SAMPLE INFORMATION:

Type	Transfusion products <ul style="list-style-type: none"> • Red blood cells • Platelets • Other product remaining after transfusion reaction was detected
Source	Blood product bag
Stability	Transport remaining blood product to the laboratory immediately after transfusion reaction is detected.
Storage Requirements	Refrigerated
Criteria for rejection	<ol style="list-style-type: none"> 1. Improperly collected, labeled, transported or handled specimens should be processed. Waiver of responsibility form SCM40110 needs to be filled out by the responsible nurse. 2. Leaking specimens should be processed, but alert the physician of the possibility of contamination.

NOTE:

- Blood products need to be processed as both body fluid culture received in blood culture vials (CXFLD →Source →blood product) and as a body fluid (CXFLD →Source: →blood product).

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REAGENTS and/or MEDIA:

- Blood agar (BA), Chocolate agar (CHO), Brucella agar (BRU) and Thioglycollate broth (THIO)
- BACTEC Plus aerobic/F culture vials and BACTEC Plus lytic /10 anaerobic culture vials
- Identification reagents: catalase, oxidase, Staph latex test, Strep latex test, etc.

SUPPLIES:

- Disposable inoculation needles
- Microscope slides
- Biosafety cabinet
- Sterile red top vacutainer tube
- Vitek 2 and supplies
- Anaerobic jar and pouch
- 35° ambient air and 35° CO₂ incubators
- Wooden sticks

SPECIAL SAFETY PRECAUTIONS:

Containment Level 2 facilities, equipment, and operational practices for work involving infectious or potential infectious materials or cultures.

- Lab gown must be worn when performing activities with potential pathogens.
- Gloves must be worn when direct skin contact with infected materials is unavoidable.
- Eye protection must be used when there is a known or potential risk of exposure of splashes.
- All procedures that may produce aerosols, or involve high concentrations or large volumes should be conducted in a biological safety cabinet (BSC).
- The use of needles, syringes and other sharp objects should be strictly limited.

All patient specimens are assumed to be potentially infectious. Universal precautions must be followed. Since viable micro-organisms are used, all cultures must be handled with appropriate precautions. All equipment in contact with cultures should be decontaminated by appropriate methods.

QUALITY CONTROL:

- Refer to Test Manual for reagent quality control procedures.

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PROCEDURE INSTRUCTIONS:

Step	Action
Processing products for blood product culture	
1	Perform all processing of blood products in the biosafety cabinet.
2	20 mL of blood product is needed for the inoculation of blood culture vials. If sufficient volume is received, proceed to step 3. If sufficient volume is not received, aseptically inject 10 to 20 mL of Thioglycollate broth into the blood product bag and mix.
3	On the aerobic and anaerobic blood culture vials place a mark at 10 mL above the level of the broth. Remove the caps from the blood culture vials and clean the septum with alcohol wipes.
4	Inspect the blood product bag and tubing and determine where the material will be taken from. Use an alcohol wipe to clean the area where the needle will be inserted.
5	Using a butterfly needle and a vacutainer barrel, aseptically insert the needle end into the blood product bag. Using the barrel, attach a blood culture bottle and allow to fill to the 10 mL mark. Repeat with the second bottle. Also collect a red top tube.
6	Remove the butterfly needle from the blood product and dispose of carefully into the sharps container. Place a piece of tape over the hole and place the blood product bag into a large biohazard bag and store in refrigerator until testing is complete.
7	From the red top tube, use a sterile pipette to inoculate Blood agar, Chocolate agar and Brucella agar and make a gram stain with one drop of the blood product. Streak for isolated growth using a disposable inoculation needle. Streak out to cover the whole plate.
8	Place BA and CHO plates in the CO ₂ incubator. Load blood culture vials into the BACTEC instrument.
9	Place BRU in anaerobic jar with anaerobic pouch as soon as practical after inoculation. Label jar with date of 48 hour read. Anaerobes should not be exposed to air for 42-48 hours after inoculation.
10	Allow smear to dry and perform Gram Stain. Gram stain must be read before culture plates. Refer to MIC20115 – Gram Stain Procedure.
11	Immediately phone results of any positive stain results for microorganisms to ordering location and document the conversation within the LIS. As well, notify the transfusion department of the results.

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12	Examine aerobic plates after 24 hour incubation. Record observations in the LIS.
13	Re-incubate CO ₂ plates for an additional 48 hours.
14	At 48 hours, examine plates and record observations in the LIS.
15	At 72 hours, examine plates and record observations in the LIS.
16	Examine anaerobic plates after 48 hours incubation and record observations in the LIS. Re-incubate BRUC anaerobically for an additional 72 hours. After 5 days, examine plate and record observations in the LIS.

INTERPRETATION OF RESULTS:

Step	Action
Interpretation of blood product culture	
1	Process blood culture vials as per MIC34000 – Blood Culture.
2	Confirm gram stain has been read prior to reading culture plates. Ensure growth on culture media correlates with gram stain results. If discordant results are found: <ul style="list-style-type: none"> • Re-examine smear and culture plates. • Check for anaerobic growth. • Re-incubate culture to resolve. • May need to inoculate special selective media. • Consider re-smearing or re-planting specimen to exclude the possibility of error.
3	Observe aerobic plates at 24 hours, 48 hours and 72 hours for growth.
4	Any growth is considered significant. Full identification and susceptibility testing as per ASTM needs to be performed.
5	If growth is present on BRU plate: <ul style="list-style-type: none"> • If growth is same as aerobic growth, re-incubate BRU for anaerobic growth for an additional 72 hours. • If growth does not resemble growth on aerobic plates, perform gram stain and aerotolerance test. Refer to MIC52600 – Aerotolerance Test.

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REPORTING RESULTS:

IF	REPORT
No growth on aerobic media after 3 days	<p>INTERIM:</p> <ul style="list-style-type: none"> Report: “No growth aerobically after 3 days” Report: “@Anaerobic culture to follow”
No growth on anaerobic media after 5 days	<p>FINAL:</p> <ul style="list-style-type: none"> Report: “No anaerobes isolated after 5 days”
Any aerobic growth	<ul style="list-style-type: none"> Report organism identification. List quantitation. Report susceptibility results as per ASTM. All growth is considered a critical result and needs to be phoned to the patient’s location. Refer to Reportable Diseases – Public Health Act as of September 2009 for reporting to HPU1. Refer to MIC35100 – Nosocomial Infection Notification Job Aid to determine if organism needs to be copied to Infection Control. Print a copy of the final report and deliver to the transfusion department to be added to the transfusion reaction work up. Freeze isolate and log into stored isolates binder.
Any anaerobic growth	<ul style="list-style-type: none"> Report organism based on gram stain results under the isolates tab. List quantitation. Add isolate comment &REF4 to state: “This organism has been referred for further identification and susceptibility testing.” Refer organism to DynaLIFE for identification and susceptibility testing as per MIC10510 – Referral of Category B Specimens to DynaLIFE. Use anaerobic transport media. Freeze isolate and log into stored isolates binder.

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LIMITATIONS:

1. False-positive results may result from contamination of the blood product at time of performing the culture.
2. False-negative results may be caused by low numbers of organism or by the fastidious nature of the infective organism.

REFERENCES:

- Jorgensen J.H., Pfaller M.A., Carroll K.C., Funke G., Landry M.L., Richter S.S., Warnock D.W. 2015. Manual of Clinical Microbiology, 11th edition, ASM Press, Washington, D.C.
- University Health Network – Mount Sinai Hospital Microbiology Department Policy and Procedure Manual: M:\SFLD\01\v18, revision date: 2015-05-26

REVISION HISTORY:

REVISION	DATE	Description of Change	REQUESTED BY
1.0		Initial Release	L. Steven

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