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| Bone Marrow Culture |
| **Purpose** | This procedure provides instructions for BONE MARROW CULTURE for the microbiology laboratory. |
| **Policy Statements** | This procedure applies to microbiologists who perform culture set-up and plate reading. |
| Principle and Clinical Significance | Bone marrow is typically aspirated from the interstitium of the iliac crest. Usually, this material is not processed for routine bacterial culture, because cultures of blood are equally as useful. Bone marrow aspirate cultures may be helpful in identifying disseminated fungal and mycobacterial infections.  |
| **Test Code** | BMC |
| **Materials** | **Reagents** | **Supplies** | **Equipment** | **Media** |
|  | * Acridine Orange Stain
* Gram Stain reagents
 | * 70% alcohol pad
* Blood transfer device or 18 gauge needle
* 1.0 mL syringes
 | * BactecTM FX blood culture instrument
 | * BactecTM Peds Plus/F aerobic medium, pink cap (BPNK)
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| Sample | * Acceptable specimen
* Bone marrow: 3 mL (minimum: 0.5 mL) placed into Bactec Peds Plus/F aerobic medium.
* For additional information: Refer to [Bone Marrow Culture](https://www.childrensmn.org/References/Lab/microbioviral/bone-marrow-culture.pdf) (Lab Test Directory) for collection, transport, storage, and rejection instructions.

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| **Special Safety Precautions** | Microbiologists/virologists are subject to occupational risks associated with specimen handling. Refer to the safety policies**:**1. [*Biohazard Containment*](https://starnet.childrenshc.org/References/labsop/mcvi/safety/mcvi-3.1-biohazard-containment.pdf)
2. [*Safety in the Microbiology/Virology Laboratory*](https://starnet.childrenshc.org/References/labsop/mcvi/safety/mcvi-3.2-safety-in-the-microbiology-lab.pdf)
* [*Biohazardous Spills*](https://starnet.childrenshc.org/References/labsop/mcvi/safety/mcvi-3.4-biohazardous-spills.pdf)
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| **Procedure** | 1. **Specimen processing**
2. Remove the plastic cap from the bottle.
3. Cleanse the stopper with 70% alcohol and allow to dry.
4. Prepare puncture site as for surgical incision.
5. Collect 0.5 – 3.0 ml of bone marrow.
6. **Inoculation**
7. Label bottle properly with the patient’s barcode label containing the name, collection date and specimen accession number.
8. Aseptically inoculate the Bactec™ Peds Plus/F bottle using a blood culture transfer device or attach an 18-gauge needle to the collection syringe.
9. **Incubation**
10. Place the Bactec™Peds Plus/F bottle in the Bactec™ FX blood culture instrument using the barcode entry.
11. Incubate for 5 days.
12. **Culture examination**
	1. BACTEC™ bottle
13. Perform subculture (CHOC, SB) and Gram stain if positive.
14. Select and inoculate additional media based on the Gram stain result.
15. If gram shows GNR, add CNA/MAC. If gram shows yeast, add SAB, CCAN.
16. If the Gram stain is negative, perform acridine orange (AO) stain.
17. If the AO is negative, do blind subcultures and continue to incubate Bactec™ bottles for 5 days.
	1. Day #1 (Positive Cultures)
18. Examine aerobic plates.
19. Gram stain each colony type.
20. Set up definitive biochemical or identification procedures on well-isolated colonies.
21. Perform antimicrobial susceptibility testing on well-isolated colonies.
22. Subculture organisms that are not well isolated to appropriate media for further work-up.
23. Reincubate primary plates and subcultures for an additional day.
24. Report preliminary results.
25. Day #2 (Positive Cultures)
26. Examine plates from the previous day for additional microorganisms.
27. Read and record identification tests and susceptibilities from the previous day.
28. Set up additional tests as needed.
29. Send updated or final report. Call critical or epidemiological important results.
30. Save a representative primary plate, whether a complete work-up was performed or not, at room temperature for 14 days in the red save boxes in case a physician calls for further studies.
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| **Method Performance Specifications** | 1. Routine bone marrow bacterial cultures are rarely useful.
2. Direct Gram stain of bone marrow is not helpful and should not be done routinely.
3. Emerging infections program (EIP): Send invasive pathogens to MDH cultured from sterile sites.

Invasive Pathogens: *Neisseria meningitidis* Haemophilus influenzaeBeta *streptococcus* group ABeta *streptococcus* group B *Streptococcus pneumoniae* *Kingella kingae* |
| **Result Reporting** | 1. Critical Value: Report all positive cultures immediately by phone to the physician or patient’s nurse. Document in the computer, the person called and the date/time of the call.
2. **No growth** cultures: Update no growth (NG) cultures daily (Desk 2 tech) in *Microbiology Automatic No-Growth Result Entry* up to 5 days.
* Enter worksheet BC and BC2 in worksheet box.
* Click ‘Add’.
* Click ‘Start Update’.
1. **Positive cultures**: Record culture results and culture work-ups in Sunquest MRE *Culture Entry* tab in Observations or Workups.

Observations: 1. GRAM POSITIVE COCCI IN CLUSTERS BEING ISOLATED AND IDENTIFIEDWorkups: Workup # 1 Workup Components Med : BPNK SC : CHOC SB  Desc : POS GMS : STPH Id : STSP1. Review Culture Summary for accuracy before filing report.
2. If growth should occur or additional testing should be requested after the culture has been finalized, remove the final status and send out a supplementary report. The code SRPT (supplementary report) must be used in SREQ or *Culture Observations* as follows:
* Updated or new culture information: In the *Culture Entry* tab, enter SRPT on an observation line followed by new results.
* Requests for additional testing: In the *Misc. Updates* tab, enter SRPT in SREQ followed by the request.
* Re-final the culture when identifications and/or testing are complete.
1. If a culture requires a correction, the code **CORR** (corrected report) must be reported on an observation line in the *Direct Exam* or *Culture Entry* tab. Refer to the procedure [MCVI 5.1 Labeling Errors/Specimen Mix-ups and Correcting Patient Data](https://starnet.childrenshc.org/References/labsop/mcvi/comp/mcvi-5.1-labeling-errors-specimen-mix-up.pdf).
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| **References** | 1. Versalovic, James. Et al, *Manual of Clinical Microbiology*, 2011, ASM press, American Society for Microbiology, Washington, D.C., pg. 294 and 305.
2. Forbes, B.A., et al., Bailey & Scott’s *Diagnostic Microbiology*, twelfth edition, Mosby, Inc., St. Louis, MO., pg. 908.
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| **Training Plan/ Competency Assessment** | **Training Plan** | **Initial Competency Assessment** |
| 1. Employee must read the procedure
2. Employee will observe trainer performing the procedure.
3. Employee will demonstrate the ability to perform procedure, record results and document corrective action after instruction by the trainer.
 | Direct observation |
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| **Historical Record** | **Version** | **Written/Revised by:** | **Effective Date:** | **Summary of Revisions** |
| 1.0 | Pat Ackerman | 1973 | Initial Version |
| 1.1 | Pat Ackerman | 01/1992 |  |
| 1.2 | Pat Ackerman | 07/07/2003 |  |
| 1.3 | Pat Ackerman | 06/02/2007 | Updated Sunquest 6.2 Reporting information. Emerging infections program (EIP): Send invasive pathogens to MDH cultured from sterile sites. |
|  | 1.4 | Jessica Craig | 05/29/2010 | Updated into online format. Changed incubation time for bottles from 7 days to 5 days. Added Bactec™ FX to Bactec™ 9240 |  |  |
| 2 | Becky Carlson | 4/14/2015 | Re-numbered from MC 407 for CMS formatting |
|  | 3 | Susan DeMeyere | 10/31/2018 | Removed Bactec™9240 blood culture instrument. Removed anaerobic culturing.  |