

ISOLATOR BLOOD CULTURES

I. CENTRAL PROCESSING AREA (CPA OR CRA) RESPONSIBILITIES:

The following applies to the Clinical Mycology Laboratory.

A. Media

1. Storage - Media used for fungus culture should be stored in the refrigerator.
2. Inoculation
 - a. Streak sediment from the Isolator tube onto one plate of BHI+GC+10% blood (BGC) and Brain-Heart Infusion Agar (BHI). This media can be replaced with IMA and Chocolate.
 - b. Tape plate lid to bottom.
 - c. The CPA technologist will process and inoculate the plates for the Mycology section.
 - d. A plate (BGC) with olive oil will be added to all blood cultures from pediatric or nursery patients by placing olive oil (2-3 drops) on the specimen and streaking together. This is to increase the speed of recovery of *Malassezia furfur* from blood. Refer to the Clinical Specimen section for additional information.
3. Incubation - Place inoculated plates of media in the plexi-glass racks marked "Mycology/AFB Isolator" and incubate at 35° in the incubator located in the AFB Laboratory.
4. Transport - Mycology personnel will pick up the inoculated media, requisitions and extra labels. This should occur at approximately 11:00 am each day.

II. RECEIPT OF INOCULATED IMA AND BHI MEDIA BY MYCOLOGY LABORATORY:

- A. Plates will be delivered as outlined in Section I-A-4..
- B. Requisitions will consist of the white copy with a label applied on the front. Two additional labels will be attached to the requisition. One of the extra labels will be placed in the log book.

III. INCUBATION OF MEDIA IN MYCOLOGY-AFB LABORATORY:

BGC and BHI Plates:

- A. Seal the IMA and BHI plates with gas permeable tape.
- B. Incubate the plates in a 35°C incubator for four weeks.

IV. READING CULTURES:

A. Plates

1. Read plates once daily for the first three days.
2. Read all plates twice a week for the remainder of the four-week incubation period (Mondays and Thursdays).
3. Discard plates after fourth week (Mondays).

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IV. READING CULTURES:

4. POSITIVE CULTURES:

- a. On the computer, as a text comment, record the number of colonies and their morphology, both macroscopic and microscopic.
- b. Record whether colonies appear to be on or off the inoculation streaks.
- c. Open and manipulate plates only within the biological safety cabinet. Use caution: Current data shows that *M. tuberculosis*, *M. avium* complex, and other species of *Mycobacterium* will grow on most mycological media..
- d. All mycobacteria will be given to the AFB technologist for identification and possible drug testing.
- e. All fungi and aerobic actinomycetes will be given to the mycology technologist for identification and possible drug testing.
- f. Immediately contact physician by phone to report positive results. If ordering physician cannot be contacted, contact Infectious Diseases.

V. INTERLABORATORY COORDINATION OF ISOLATES:

A. Bacteria Recovered by Mycology

1. Prepare a subculture using a Sheep blood agar plate.
2. Give the subculture to the bacteriology technologist working up that patient.
3. The culture containing either the final identification, susceptibility testing data, or both is to be returned to the mycology supervisor after the technologist in Bacteriology has entered the data in the computer without finalizing results.

B. Fungi Recovered by Bacteriology

1. The bacteriology technologist will prepare a subculture of a yeast using a Sabouraud glucose agar tube. If a mould is recovered, the plate is sealed with scotch tape and then given to the mycology technologist.
2. The subculture or original plate and bacteriology work card are given to the mycology technologist.
3. The mycology technologist will place the final identification of the fungus on the bacteriology culture screen.

VI. REPORTS:

A. Bacteria Recovered by Mycology

1. Final identification from bacteriology will be entered into the computer by the bacteriology technologist.
2. A bacteriology identification charge, and susceptibility testing charge, if appropriate, will be entered by the bacteriology technologist.

B. Fungi Recovered by Bacteriology

1. Final results from mycology will be entered into the computer by the mycology technologist.
2. A mycology identification charge will be entered by the mycology technologist.