

## MICROBIOLOGY

### GENERAL CONSIDERATIONS:

1. The Microbiology laboratory is open 5 days a week from 8 AM to 10:00 PM. And on Sturday and Sunday. Stat requests after these times should be delivered to the Stat Laboratory.
2. **STAT** requests are available at any time. Specimens must be handed to a technologist in the MICROBIOLOGY laboratory before 4:30. After 4:30 PM Saturday and Sunday and 10PM Monday through Friday, specimens must be handed to a technologist in the STAT laboratory.
3. Specimens must be properly labeled and received in sterile containers in order to be processed. Minimum information necessary is **patient name, identification number, Doctor ordering culture or room number, date of collection and specimen type**. Sterile containers and transport media are available from purchasing. Transport media is Amies with charcoal which will support the growth of Neisseria gonorrhoeae.
4. Improperly labeled or unlabeled specimens are unacceptable for culture. The only exceptions will be for those specimens requiring an invasive procedure or instrumentation for collection. In these instances, the proper label must be brought down and verified **in person by the nurse or physician**. All changes must be initialed.
5. Specimens must be sent in tightly sealed containers with no external spillage. Leaky contaminated containers are a hazard to personnel and will not be accepted. If a container becomes contaminated during collection, wipe off with disinfectant before transporting to the laboratory.
6. Disinfect specimen containers from isolation units with disinfectant and place in appropriate bag for transportation. Keep specimen containers upright.
7. Specimens for culture must be brought to the laboratory promptly after collection. If delay is unavoidable, they may be kept in the refrigerator for up to 24 hours with the following exceptions:
  - a. CSF must **not** be refrigerated as cold temperatures may destroy some organisms that may cause meningitis. Deliver to the laboratory **and HAND TO A TECHNOLOGIST**.
  - b. Specimens for anaerobes should be left at room temperature if collected in the BBL Anaerobe Transport System – Port-a-Cult (available in purchasing)
8. Preliminary reports are available for routine cultures in 24 hours, 48 hours for anaerobic cultures, 48 hours for acid fast cultures and within 7 days for mycology. All smears, including KOH, wet preps, India Inks, acid fast smears and gram stains are reported in 24 hours.

### ACRIDINE ORANGE STAIN

**Availability:** 8:00 AM - 4:00 PM Monday-Friday in Microbiology only

**Collection:** Material to be stained is submitted in a sterile container, can also be submitted on a clear (unfrosted) slide.

## **ACID FAST BACILLI (AFB OR MYCOBACTERIA)**

**Note:** For direct detection of AFB (*M. tuberculosis*) by PCR see under "Mycobacterium Tuberculosis, Direct Detection from Specimen".

**Availability:** Specimens are processed once a day between 11 and 12:00  
Specimens received after this time will be held over for processing until the following day.

**Collection: SPUTUM:**

1. 10 ml of first morning sputum or induced sputum (can be induced by inhalation of aerosolized 10% sodium chloride solution).
2. A series of 3-5 single early morning specimens, collected on successive days is recommended.
3. 24 hour specimens are **NOT ACCEPTABLE**.

**BRONCHIAL WASHINGS:** Collect in sterile container.

**GASTRIC WASHING:**

1. Collect a minimum of 50 ml of an early morning specimen in a sterile container.
2. Food, drink, or oral medications should not be given after midnight.
3. Deliver **immediately** to the laboratory.

**BLOOD:**

1. See **BLOOD CULTURES** for collection.
2. Inoculate one vial of MB BactiAlert MP medium available from phlebotomy.
3. Blood cultures should be taken to the laboratory as soon as possible.

**BODY FLUIDS:**

1. Collect fluid using sterile technique. See **MICROBIOLOGY – BLOOD CULTURES** for removal of skin contaminants.
2. Fluid should be placed in a sterile container or left in the syringe. If leaving fluid in syringe, remove the needle using appropriate safety device and technique and replace syringe cap.
3. Swabs are unacceptable specimens for fluid.

**FECES:** Collect in sterile container.

**TISSUE:** Collect in sterile container. Do NOT add formalin or wrap in tissue paper or gauze. If tissue is very small, add a small amount of sterile normal saline.

**URINE:**

1. Collect approximately 50 ml of clean voided early morning urine in a sterile container.
2. 24 hour urine specimens are unacceptable.
3. A series of 3 single early morning specimens collected on

successive days is recommended.

**Method:** **MB BactiAlert** positive cultures are sent to the State Health Department Laboratories for identification by DNA probes for M. tuberculosis, M.avium complex, M. gordonae and M.kansasii. 2-7 day turn-around-time. Other non-tuberculosis Mycobacteria are identified by conventional means.

Negative cultures

**Note:** For direct identification of MTB from the specimen, see Mycobacterium Tuberculosis.

**AMOEBAS:** See PARASITES, AMOEBAS

### **BLOOD CULTURES:**

**Availability:** Blood cultures are collected by the phlebotomy staff from 6AM to 12 midnight seven days a week. It is the responsibility of the doctors, I.V. nurse or other hospital personnel to collect blood cultures from midnight to 6 AM with the following exceptions:

1. MSICU, CSU, and ED will draw their own blood after 9 PM.
2. Pediatrics will draw blood on patients under 5 years of age.

**Specimen:** Blood collected in the following blood culture media:

Aerobic Standard Media	Blue Top	5-10ml
Anaerobic Standard Media	Purple Top	5-10 ml
FAN Aerobic Media	Light Green Top	5-10 ml
Pediatric/Low Volume Media	Yellow Top	0.5-4 ml
Mycobacteria (AFB) BactiAlert MB	Black Top	3-5 ml
Fungus – FAN Media	Light Green Top	5-10 ml

**Collection:** ***Routine Blood Cultures*** – one **aerobic** and **anaerobic standard media**.  
***Patients on antimicrobial therapy*** – one **aerobic FAN** and one standard **anaerobic bottle**.

***Pediatrics or short draw***– one **Ped Bact T medium**

***Fungal Culture*** – one **FAN aerobic medium** and one standard **anaerobic media**.

***AFB or Mycobacteria*** – one **BactiAlert MB**

1. Ask the patient their name and date of birth. Check against the arm band and the patient label.
2. Remove the flip off cap from the BactAlert vial and clean the rubber seal with a 70% alcohol prep. **DO NOT USE IODINE TO CLEAN BOTTLE TOPS as this may cause deterioration of the rubber stoppers,**
3. After choosing the puncture site, **prepare the skin** using Chloroprep. Scrub area back and forth for a period of 30 seconds. Let dry on the skin for 30 seconds.

4. Using standard precautions, with the bevel of the needle up, draw 0.5 to 10 ml of blood depending on the number and type of bottles to be inoculated. Optimal volume of blood for BactiAlert bottles is 10 ml. **It is very important that you do not overfill the vials as this will cause false positive results. Pediatric bottles should receive 0.5 to 4 ml of blood. Do not overfill.**
5. Place a sterile gauze pad over the puncture site while the needle is withdrawn and hold firmly until all bleeding is stopped.
6. Place a name label on each of the bottles without obstructing the bottle bar code label or bottom of bottle (sensor) and initial the bottle with the Mediatech initials.
7. Take blood culture bottles to the specimen processing area of the laboratory.

**Methodology:** BactiAlert Organon Teknika

**Notes:** Turn around time for negative cultures is 5 days. Positive cultures will be called to the nursing unit/doctor for outpatients or the hospitalist on call.

**BORDETELLA PERTUSSIS/PARAPERTUSSIS CULTURE:** See CULTURE;BORDETELLA PERTUSSIS/ PARAPERTUSSIS

**BORDETELLA PERTUSSIS/PARAPERTUSSIS SMEAR BY FA:**

**Availability:** Specimens are sent to a reference laboratory Monday through 9:30 AM Friday. Specimens received after 9:30 AM Friday will be held until Monday. Special arrangements can be made for later delivery on Friday up till 3:00PM.. The Md State Health Department is closed on weekends.

**Specimen:** Two air-dried clear (unfrosted) slides of nasopharyngeal secretions.

**Method:** Direct Immunofluorescence. Turn around time 1-3 days.

**Comments:** The kit for collection of specimens is available from the micro Laboratory. It contains slides for smears and transport media for The culture. It is not necessary to order both. See also "Culture Bordetella". **If the specimen cannot be transported the same day, place slides in freezer, transport tube in 35°C incubator.**

**CLOSTRIDIUM DIFFICILE TOXIN A ASSAY:**

**Availability:** Test is performed once a day in the afternoon at about 1:00PM. Specimens received after that time will be held over to the following day.

**Specimen:** 5 grams (5 ml.) of *unformed loose, or watery stool* in sterile

container. May be refrigerated for up to 48 hours.

**Comments:** **FORMED STOOLS ARE UNACCEPTABLE.**

**Method:** Monoclonal-based enzyme immunoassay for rapid detection of *C. difficile* toxin A.

**CRYPTOSPORIDIUM:** See PARASITES: CRYPTOSPORIDIUM

**CYCLOSPORA:** See PARASITES: CYCLOSPORA

**CULTURE - ACTINOMYCES:** See Culture – Anaerobic Bacteria

**CULTURE - ANAEROBIC BACTERIA:**

**Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**

**Specimens:**

1. Transtracheal aspirate
2. Urine: submit suprapubic tap, nephrostomy tube, suprapubic catheter
3. Sterile body fluids
4. Exudates, aspirated pus from deep abscesses or wounds
5. Surgical specimens
6. Cul-de-sac aspiration, culdocentesis, placenta, septic abortion
7. Prostatic or seminal fluid.

**SWABS:**

1. Place swab in Port-a-cult Anaerobic Transport System Available from purchasing.
2. Insert swab to within approximately 5 mm from bottom of tube; break off handle. **DO NOT USE** if medium has turned pink.
3. **DO NOT REFRIGERATE.** Leave on counter in laboratory.

**FLUIDS/ASPIRATES:**

1. Follow standard procedure to obtain specimen. Disinfect superficial area using technique described in **MICROBIOLOGY - BLOOD CULTURES # 5** to eliminate contamination with normal skin flora.
2. Place specimen in sterile container or leave in syringe. If the specimen has been aspirated by syringe it is best to leave it in the syringe. Remove the needle using appropriate safety device and technique and replace syringe cap.

**TISSUE:** Place in sterile container. Do NOT add formalin or wrap in paper or gauze.

**Comments:** *Anaerobic cultures WILL NOT BE DONE on the following*

***specimens:***

1. Nose, throat;
2. Sputum, saliva or bronchoscopy aspirates;
3. Feces, gastric contents;
4. Voided or catheterized urines; Anaerobic culture will be done on suprapubic taps only;
5. Colostomy or ileostomy effluent or sites;
6. Superficial skin wounds and lesions;
7. Ears (outer).

**CULTURE - ASCITIC FLUID (PARACENTESIS):** See CULTURE: FLUIDS

**CULTURE- BLOOD: see under BLOOD CULTURE**

**CULTURE - BORDETELLA PERTUSSIS:**

**Availability:** Specimens are sent to a reference laboratory Monday through 9:30 AM. Specimens received after 9:30 AM on Friday will be held over till Monday. Special arrangements can be made for later delivery on Friday up to 3:00PM. The Md. State Health Department is closed on weekends.

**Specimen:** **Nasopharyngeal swab** in Regan-Lowe Transport media. Stable for 72 hours. Hold at room temperature. Contact microbiology for transport media. If received in routine transport media, good for 24 hours refrigerated.

Bronchial aspirations: hold in original container; refrigerate.

**Method:** Turn around time is 7-10 days.

**Comment:** Direct immunofluorescence examination of a smear is available as a separate test. See **BORDETELLA SMEAR BY FA**. The Collection kit contains both slides for smear and transport for culture. It is not necessary to order both. Hold transport tube in 35°C incubator if cannot transport same day, (See Bordetella smear)

**CULTURE: CATHETER TIP (INTRAVENOUS):**

**Availability:** See **MICROBIOLOGY- GENERAL CONSIDERATIONS**

**Specimen:** Catheter tip

**Collection:**

1. Decontaminate skin as indicated for blood culture.
2. Use sterile forceps to remove catheter.
3. Submit tip in sterile container.

**CULTURE, CERVICAL FOR ROUTINE OR GC:** See CULTURE: GENITAL

### **CULTURE, C.S.F.:**

- Availability:** 24 hours, 7 days a week
- Specimen:** Spinal fluid in sterile container. Swabs are NOT acceptable.
- Special Instructions:**
1. Specimens must be transported to the laboratory IMMEDIATELY and handed to a TECHNOLOGIST. DO NOT place in the refrigerator.
  2. Tube # 2 should be used for culture if available, unless otherwise stated by the physician.
  3. Culture should be done if the cell count is  $\geq 10$ .
- Comments:**
1. Direct smears are done routinely and results called to physicians and nursing units.
  2. Cultures are done on all CSF with cell counts equal to or greater than 10.

### **CULTURE - DIPHTHERIA:**

- Availability:** Specimens are sent to a reference laboratory Monday through 9:30 AM Friday. Specimens received after 9:30 on Friday will be held until the following Monday.. Special arrangements can be made for later delivery on Friday up until 3:00PM. The Md State Health department is closed on weekends..
- Specimen:** Nasopharyngeal swab in transport media and a portion of the membrane, if present.
- Comment:** Turn around time is 3-6 days.

### **CULTURE, EYE:**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**
- Specimen:** Swab in transport media.

### **CULTURE - FLUIDS; JOINT, PLEURAL, ASCITIC (PARACENTESIS):**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**
- Specimen:** Body fluids may be submitted in a sterile container or the syringe used in obtaining the fluid. Specimens submitted in the syringe must have the needle removed using the appropriate safety device and technique. Replace syringe cap. *EDTA, Heparin, Citrate are unacceptable.*
- Collection:** Body fluids are usually collected via percutaneous needle aspiration. It is essential that the overlying skin be disinfected

thoroughly before the aspiration is performed and that the collection procedure itself be done by strict aseptic technique. See - **MICROBIOLOGY - BLOOD CULTURES** for proper skin preparation.

**CULTURE: GC:** See CULTURE: NEISSERIA GONORRHEA

**CULTURE: GENITAL:**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**
- Specimen:** Cervical, endocervical, urethral, vaginal, penile swab in transport medium.
- Collection:** For vaginal, cervical and endocervical specimens should be collected using a bivalve speculum moistened with water. Soap and jelly lubricants should not be used.
- Comment:**
1. Routine cultures are always screened for GC; therefore, if both are requested, order only routine culture.
  2. Requests for GC smears should always have a culture taken for confirmation.

**CULTURE, FUNGI:**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**
- Specimen:** ***SPECIMEN FOR DERMATOPHYTES:***
1. **HAIR:** hair may be removed with forceps or a disposable brush or comb.
  2. **SKIN:** cleanse area with an alcohol wipe; remove epidermal scales at active border of lesion with a sterile scalpel.
  3. **NAILS:** cleanse area with an alcohol wipe. Scrape outermost layer with sterile scalpel. Deeper scrapings, debris from under edges of nail and nail clippings are suitable.
  4. Place specimen in clean container such as envelope, petri dish, or sterile urine/sputum cup. Do NOT PLACE in Transport Media.
- URINE:** Same as routine bacterial culture.
- SPUTUM:** First morning specimen is recommended. Teeth should be brushed extensively first or dentures removed. Rinse mouth with mouthwash and several washes of sterile water or saline. Nasopharyngeal secretions and saliva are unsatisfactory.

***SUBCUTANEOUS TISSUES, FLUIDS AND BIOPSIES:***

Collect the same as for routine culture. Send to the lab in a sterile container to be processed immediately before bacterial and autolytic processes degrade the specimen. DO NOT REFRIGERATE A SPINAL FLUID.

**SWABS:** Place in routine transport media.

**BLOOD:** See **BLOOD CULTURE**

**Comment:** Negative cultures are held for 4 weeks, negative tissue cultures are held for 6 weeks.

**CULTURE - JOINT FLUID:** (SEE "CULTURE - FLUIDS")

**CULTURE - LEPTOSPIRA - URINE OR BLOOD:**

**Availability:** 8:00 AM-3:30 PM Monday through Friday

**Specimen:** Urine, CSF, blood

**Collection:** **BLOOD:** 10 ml of heparinized (green-top) whole blood. Hold at room temperature. Send to Laboratory.

**URINE:** After one week of the disease, collect 3 to 4 specimens at least a day apart. Send to laboratory immediately. (In the lab, place 2 to 3 drops of urine in the Leptospira transport media (Fletcher's medium) available in Microbiology. Hold at room temperature.)

**CSF:** Send to laboratory immediately. (In the laboratory, place 3 to 5 drops of spinal fluid in the transport medium. Hold at room temperature. Send to Maryland State Health Department.)

**Notes:**

1. Collect specimens before administering antibiotics, if possible.
2. Do not use contaminated (cloudy) transport medium.
3. Do not use transport medium after the expiration date.

**Comments:** Send to Maryland State Health Department. Closed weekends and holidays. Most pathogenic strains are detected after 6 to 14 days of incubation; however they may take as long as 3 to 4 months.

**CULTURE: MONILIA:** - See CULTURE: FUNG

**CULTURE- NASOPHARYNGEAL (N-P):**

<b>Availability:</b>	See <b>MICROBIOLOGY - GENERAL CONSIDERATIONS</b>
<b>Specimen:</b>	Calcium Alginate swab on flexible wire. (Available from purchasing)
<b>Collection:</b>	Pass a dacron, cotton or calcium alginate swab on a flexible wire through nostrils into the nasopharynx being careful NOT to touch the nasal mucosa. Rotate swab and remove. Place in transport medium for delivery to the laboratory.

**CULTURE, NEISSERIA GONORRHEA (GC):**

<b>Availability:</b>	See <b>MICROBIOLOGY - GENERAL CONSIDERATIONS</b>
<b>Specimen:</b>	Body fluids, blood, prostatic fluid, eye, rectal, vaginal cervical, throat
<b>Collection:</b>	Submit fluids in sterile containers, swabs in transport medium.
<b>Comments:</b>	Smears submitted for GC should have culture taken for confirmation.

**CULTURE, NOSE:**

<b>Availability:</b>	See <b>MICROBIOLOGY - GENERAL CONSIDERATIONS</b>
<b>Specimen:</b>	Sterile swab in transport media.
<b>Collection:</b>	Swab nasal mucosa. Swabs may be moistened with sterile saline. Place swab in transport media.

**CULTURE, PENILE:** See CULTURE, GENITAL

**CULTURE, PLEURAL FLUID:** See "CULTURE, FLUIDS

**CULTURE - STOOL OR RECTAL SWAB FOR ENTERIC PATHOGENS:**

<b>Availability:</b>	See <b>MICROBIOLOGY - GENERAL CONSIDERATIONS</b>
<b>Specimen:</b>	Fresh stool in sterile container or rectal swab in transport media.
<b>Collection:</b>	If swabs are used to collect specimen, they should be passed beyond the anal sphincter, carefully rotated, and withdrawn. Place in transport media.
<b>Comment:</b>	Stools are routinely screened for Salmonella, Shigella, Yersinia,

Campylobacter, Staphylococcus aureus and E. coli O157. In June, July and August they are screened additionally for Vibrio.

**CULTURE, SPUTUM:**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS.**
- Specimen:** Early morning sputum
- Collection:** Rinse mouth well with mouth wash. Instruct patient to produce a deep cough sample, not just saliva.
- Comments:** Sputa containing more than 25 epithelial cells per low power field are unacceptable for routine culture due to contamination with saliva.

**CULTURE, THROAT:**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**
- Specimen:** Throat swab in transport media
- Collection:** Obtain by direct visualization with rayon (dacron) swab. Both tonsillar areas, the posterior pharynx, and any areas of inflammation, ulceration, or exudation should be sampled. The tongue should be depressed with a tongue blade or spoon to minimize contamination of the swab with oral secretions detrimental to the quality of the specimen.

**CULTURE, THRUSH:** See CULTURE: FUNGI

**CULTURE, TISSUE:**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**
- Specimen:** Submit tissue or tissue biopsy in a sterile container. For small biopsy specimens, add small amount of **sterile saline**. Do **NOT** place in *tissue paper or gauze or transport media*. Do **NOT** add *formalin*. Specimens submitted in formalin for culture are **UNACCEPTABLE**.

**CULTURE, URETHRAL; ROUTINE AND/OR GC:**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**
- Specimen:** Swab in transport media

**CULTURE, URINE:**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**
- Specimen:** Urine; clean caught, catheter, suprapubic aspirate in a sterile container or BD Urine Culture Collection Vacutainer Tube (grey top).
- Collection:** For collection of clean caught urines, patients should be instructed in the proper use of the clean caught collection kit, and the importance of thorough cleansing. After collecting the urine, draw up into the grey topped BD Urine Culture Collection vacutainer tube. These tubes contain boric acid and sodium formate to prevent overgrowth of bacteria.
- Comment:** If unable to transport to the laboratory immediately, urine collected in a sterile container may be held in the refrigerator for up to 24 hours. Urines collected in the BD Urine Culture Collection Vacutainer tube are stable for 48 hours.
- Unacceptable Specimens:** Urines collected in sterile containers held at room temperature for over an hour and urines refrigerated for over 24 hours are unacceptable for culture.  
Urines collected in the BD Urine Culture Collection tube for more than 48 hours  
Foley catheter tips are unacceptable for culture.

**CULTURE, WOUNDS; ABSCESS, EXUDATE, DRAINAGE:**

- Availability:** See **MICROBIOLOGY - GENERAL CONSIDERATIONS**
- Specimen:** Swab in Transport Media
- Collection:**
1. Decontaminate wound surface.
  2. Swab or aspirate deep areas, not surface.
  3. Submit purulent material in a transport swab.
- Comment:** For culture of anaerobes, see **CULTURE: ANAEROBE.**

**CULTURE, VAGINAL FOR ROUTINE OR GC:** See **CULTURE: GENITAL**

**CULTURE, WHOOPING COUGH:** See **CULTURE: BORDETELLA**

**CYCLOSPORA, SEE PARASITES, CYCLOSPORA**

**DIPHTHERIA:** See **CULTURE: DIPHTHERIA**

**ENTERIC PATHOGENS:** See **CULTURE: STOOL**

**ENTEROBIUS VERMICULARIS:** See PARASITES: PINWORM

**ENTEROCYTOZOOM SPECIES:** See PARASITES: MICROSPORIDIUM.

**GC CULTURE:** See CULTURE: NEISSERIA GONORRHEA

**GROUP B STREPTOCOCCUS SCREEN:**

- Availability:** See **MICROBIOLOGY, GENERAL CONSIDERATIONS.**
- Specimen:** Cervical or vaginal/rectal swab.
- Collection:** Vaginal and cervical specimens should be collected using a bivalve speculum moistened with water. Soap and jelly lubricants should not be used.
- Comments:** Group B Strptococcus Screens are done by culture in accordance with the CDC recommendations. Turn around time is 72 hours. Rapid testing for Group B Streptococcus is no longer available.

**INDIA INK PREPARATION:**

- Availability:** 24 hours, 7 days a week
- Specimen:** CSF, body fluids, sputum, aspirates

**INFLUENZA A& B, RAPID TEST**

- Availability:** 24 hours, 7 days a week
- Specimen:**
1. Nasal washes: 2-3 ml are recommended. Excess volume may Decrease test performance.
  2. Nasal aspirates: should be collected in volumes between 0.5 And 1 ml and placed in 2-4 ml of normal saline or viral transport media.
  3. Nasal swabs. Place into 1 to 3 ml of normal saline. *Use only Rayon or dacron tipped swabs with plastic or aluminum shafts.*
- Comments:**
1. Nasal washings or aspirates are the recommended specimen.
  2. Do **not** use calcium alginate.

**KOH PREPARATION:**

- Availability:** 8:00 AM-4:00 PM, deliver to Microbiology
- Specimen:** Hair, scrapings of skin or nails. See **FUNGAL CULTURE** for collection of specimen. Swabs are unacceptable.

## **LEGIONELLA URINARY ANTIGEN:**

- Availability:** 8:00AM – 10:00PM Monday through Friday, 8 – 4 PM Saturday and Sunday.
- Specimen:** Urine collected in standard urine containers. Boric acid may be used as a preservative. Specimens may be held at room temperature for up to 24 hours. Grey top vacutainers for culture are not acceptable. (grey top vacutainers contain boric acid and sodium formate)
- Methodology:** Binax NOW Legionella Urinary Antigen Test.
- Comments:**
1. This test detects Legionella pneumophila serotype 1 only
  2. A negative result does not exclude infection with L. Pneumophila.
  3. Excretion of Legionella antigen in urine may vary, beginning as early as 3 days after onset of symptoms and persist for up to 1 year afterward.

**MICROSPORIDIUM:** See PARASITES, MICROSPORIDIUM.

## **MBC - MINIMAL BACTERICIDAL CONCENTRATION (LEVEL):**

- Availability:** Organism must be isolated in the laboratory. This test is referred to a reference laboratory.
- Specimen:** Pure culture of organism available in the laboratory.
- Comments:** Indicate antibiotic(s) to be tested. There is a charge for each antibiotic tested.

## **MIC - MINIMAL INHIBITORY CONCENTRATION:**

- Availability:** 8:00 AM to 4:00 PM, Saturday through Sunday. MIC are done routinely on pathogens from selected sites when the susceptibility pattern cannot be accurately predicted.
- Specimen:** Pure culture of the organism available in the laboratory.
- Comments:**
1. Test takes 24 hours after isolation of the organism.
  2. Indicate particular antibiotic to be run if not on routine panel of drugs.
  3. Antibiotics unavailable at this hospital will be sent to a

reference laboratory for testing.

**MYCOBACTERIA:** See ACID FAST BACILLI

**MYCOBACTERIUM TUBERCULOSIS, DIRECT IDENTIFICATION FROM SPECIMEN:**

- Availability:** Specimens are sent to the Maryland State Health Department. Available Monday through Thursday and up to 12 noon on Friday. (MTB Direct Test)
- Specimen:** Sputum, bronchial washings from high risk patients.
- Comment:** Test is available for M. tuberculosis only
- Method:** Polymerase Chain Reaction. Turn around time is 2-4 days. This test is not available on weekends.

**PARASITES, BLOOD; (MALARIA, BABESIA, FILARIASIS, LEISHMANIASIS, TRYPANOSOMIASIS):**

- Availability:** Hematology. 8:00 AM to 2:30 PM. For blood received after 2:30, smears will be made and held until the following day.
- Specimen:**
1. Fresh whole blood from puncture of earlobe or fingerstick.
  2. Venous blood collected in EDTA anticoagulant (lavender top tube or capillary blood collected in EDTA microtainer.
  3. Bone marrow aspirates or liver biopsy may be useful in the diagnosis of trypanosomiasis and leishmaniasis.
- Collection:**
1. Allow alcohol disinfectant to dry before the site is punctured or there may be fixation of erythrocytes which will interfere with the preparation and staining of thick films.
  2. The timing of the collection of blood parasites depends on the parasite disease suspected. Please consult with laboratory or pathologist.
- Method:** Giemsa stain of blood smear; thick and thin smears

**PARASITES, CRYPTOSPORIDIUM/GIARDIA OOCYSTS:**

- Availability:** 8:00 AM - 4:00 PM Monday through Friday
- Method:** Direct Fluorescent Antibody Stain
- Specimen:** Feces, colonic or duodenal aspirate

**Collection:** Submit fresh stool, colonic or duodenal aspirate in sterile container. Para-Packs are acceptable for specimens where there will be a delay in transit or over a weekend. Liquid stools should be placed in 10% formalin within one hour of collection.

**Comment:** Turn around time 24 hours Monday through Friday.

**PARASITES: CRYPTOSPORIDIUM ANTIGEN DETECTION, EIA:**

**Availability:** Sent to Reference Laboratory 8:00 AM to 4:00 PM Monday through Friday.

**Specimen:** 1 gram of fresh stool <=48 hours post collection.

**Method:** EIA

**PARASITES, COLONIC/DUODENAL ASPIRATE:**

**Availability:** 8:00 AM-4:00 PM Monday through Friday.

- Collection:**
1. Place specimen in sterile container. DELIVER TO THE MICRO LAB IMMEDIATELY.
  2. For specimens collected after 4 PM, place in parasitology kit.
  3. Specimen should be received while warm. If delay is unavoidable, keep at room temperature or refrigerate.

**Comments:** Artificial warming is contraindicated.

**PARASITES, CYCLOSPORA:**

**Availability:** Sent to Reference Laboratory 8 AM to 4 PM Monday through Friday.

**Specimen:** Approximately 2 grams of stool (or teaspoon amount) preserved in 10% formalin. Fresh stool, swabs or stool on a diaper are unacceptable.

**Method:** Modified acid-fast stain on air dried methanol fixed smears.

**PARASITES, MICROSPORIDIA: (*Enterocytozoon species*)**

**Availability:** Sent to reference laboratory 8:00AM to 4:00 PM Monday through Friday.

**Specimen:** 0.5 grams or 1 mL fresh stool or duodenal aspirate in 10% formalin, 1:3 dilution.

**Method:** Modified Trichome Stain.

## **PARASITES, NIH PREPARATION FOR PINWORMS:**

- Availability:** 8:00 AM to 4:00 PM Monday through Friday.
- Specimen:** Anal scotch tape preparation
- Collection:**
1. At one end of the strip of transparent tape, fold the tape upon itself to provide a non-adhesive area for handling.
  2. Apply the remainder of the tape to a clear glass slide extending over the end about 1 cm. on the under surface of the slide.
  3. When the sample is to be taken, hold the slide against a tongue depressor 2.3 cm. below the end and lift the long portion of tape from the surface of the slide.
  4. Loop the tape over the extended end of the depressor to expose the gummed surface. Hold the tape and slide against the depressor to provide tension and a firm support for the loop of scotch tape.
  5. Separate the buttocks of the patient and press the gummed surfaces against several areas of the perianal region.
  6. Replace the tape on the slide (to which it has remained attached on the undersurface) and smooth the tape with cotton or gauze.
- Comments:**
1. Use only clear, transparent tape and clear slides. **Frosted tape and/or slides are unacceptable.**
  2. Specimens are best obtained a few hours after the person has retired or the first thing in the morning before a bowel movement or bath.

## **PARASITES, STOOL:**

- Availability:** 8:00 AM-3:30 PM Monday through Friday. Tests are run once a day. Specimens received after this time will be held to the following day.
- Collection:** Must be sent in parasite kit. Follow instructions received with kit. Liquid stools should be placed in preservative (10% formalin) within one hour of collection.
- Comments:**
1. Contamination with urine should be avoided.
  2. Laxatives containing oil, magnesia, bismuth or barium enema interfere with examination for parasites. Specimens should not be repeated for a minimum of 3 days or until interfering substances are cleared from stool.
- Method:** Formalin-Ethylacetate (Ritchie)

**PARASITES, SIGMOIDOSCOPY:** See PARASITES, COLONIC/DUODENAL ASPIRATE

## **PARASITES, PNEUMOCYSTIS CARINII:**

**Availability:** This test is performed in CYTOLOGY. Please send specimen and request to Cytology. Specimens received after 12:30 will be held for processing the following day.

**Specimen:** PREFERRED: Open lung biopsy and transthoracic needle biopsy.  
ACCEPTABLE: Bronchial washings, bronchial brushings, bronchial alveolar lavage and sputum

#### **RAPID SCREEN FOR GROUP A BETA STREPTOCOCCUS:**

**Availability:** 8:00 AM - 4:30 PM, 7 days a week, deliver to microbiology. After 4:30 PM until 8 AM, deliver to the Stat laboratory.

**Specimen:** Throat swab *ONLY*. Other specimen types are UNACCEPTABLE.

**Collection:** Use a swab with a **dacron or rayon tip**. Do not use swabs with calcium alginate or cotton tips, or wooden swabs. Transport in Modified Stuart's medium or equivalent. Do not use transport media that contains gelatin, agar, or charcoal. Proper swabs and Transport media are available from purchasing. If there is a delay in transport to the laboratory, swabs should be refrigerated and transported within 48 hours.

#### **RAPID SCREEN FOR GROUP B BETA STREPTOCOCCUS**

**Availability:** 8:00 AM – 4:30 P, 7 days a week , deliver to microbiology. After 4:30 PM and until 8:00 AM, deliver to STAT laboratory.

**Specimen:** Vaginal/rectal swab in transport media.

**Comment:** If susceptibilities to clindamycin and erthromycin are requested, please enter into the computer as a comment.

#### **RSV, RAPID TESTING:**

**Availability:** 24 hours, 7 days a week

**Specimen:**

- Nasal washings:** 2-4 ml are recommended. Excess eash volume may decrease test performance.
- Nasal aspirates** should be collected in volumes between 0.5 and 1.0 ml and placed in 2-4 ml of viral transport medium or normal saline.
- Nasal swabs:** place in 1 to 3.0 ml of normal saline. *Use only rayon or dacron tipped swabs with plastic shafts.*

**Comments:** 1. Do not use calcium alginate swabs.

2. Nasal washings or aspirates are the recommended specimen.

**SCHLICHTER TEST (SERUM ANTIBACTERIAL TITER, SERUM CIDAL, BACTERIOCIDAL LEVEL):**

- Availability:** This test will be set up Monday through Friday. It is necessary to have a viable culture of the organism. Please call Microbiology before ordering this test.
- Specimen:** 2 ml serum ( one red-topped vacutainer); bacterial isolate.
- Collection:** The patient's blood should be drawn **immediately before** the next dose of antibiotic. This is the trough level. Collection time for the **peak level** depends upon the antibiotic and its route of administration and the time should be specified by the physician. If no times are specified, draw a trough level.

**SKIN SCRAPING FOR SCABIES:**

- Availability:** 8:00 AM - 4:00 PM Monday through Sunday.
- Specimen:** Scrapping of infected area of skin
- Collection:**
1. Supplies needed: sterile surgical blade or other sharp instrument for obtaining scrapings; mineral oil; 70% alcohol swabs; non-frosted slides, cover slips; and petri dish. ( slides, cover slips and petri dish available from Micro Lab.)
  2. Place one or two drops of mineral oil on slide.
  3. Cleanse area of skin with alcohol.
  4. Dip the surgical blade into the mineral oil on the slide and transfer a small drop of oil to the skin surface to be scrapped.
  5. Holding the blade at 33° to 45° angle, lift off (scrape) the superficial layer of skin and transfer to slide.
  6. Coverslip preparation; place in petri dish and send to the microbiology laboratory.

**SYNERGY STUDIES; SUSCEPTIBILITY, DRUG STUDY:**

- Availability:** Specimens are sent to a reference laboratory. They must be received in the microbiology laboratory before 4:30 Monday through Friday and before noon on Saturday to be sent that day. Specimens received after this time will be held until the following day. There are no send outs on Sundays.
- Specimen:** Isolated organism
- Comment:** Indicate isolate, source of isolate, and antibiotic combination(s). There is a charge for each antibiotic.

**VINCENT'S ANGINA: TRENCH MOUTH:**

<b>Availability:</b>	See <b>MICROBIOLOGY - GENERAL CONSIDERATIONS.</b>
<b>Specimen:</b>	Scrapings of infected gums
<b>Collection:</b>	Scrapings should be placed on a clean, non-frosted slide and placed in a petri dish for transportation to the microbiology laboratory. Do <b>NOT</b> use fixatives.
<b>Method:</b>	Gram stain

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