

**JOHNSON MEMORIAL HOSPITAL
LABORATORY PROCEDURE**

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| Procedure: Collection and Transport of Microbiology Specimens | Policy Number: 109 |
| Function: Microbiology Guidelines For Submission Of Specimens | Department: Microbiology |
| Formulated By: Bill Lolley | Number of Pages: 16 |

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| Medical Director Approval: | Date: | |
| Laboratory Manager Approval: | Date: | |

I. Principle:

A. The JMH Microbiology department provides the following guidelines for the collection and transport of culture specimens for microbiology studies. These guidelines are designed to provide adequate instructions to ensure proper handling and processing of patient specimens in an expeditious and safe manner before they are received by the laboratory.

Located at each nursing station is a copy of JMH Laboratory Procedures which contain relevant guidelines for collection, handling, transporting and submission of Microbiology specimens for:

1. Isolation of Bacterial and Fungal Studies
2. Serological Analysis Studies and
3. Screening for Parasitic Studies

Each member of the nursing staff is encouraged to become familiar with these guidelines to ensure proper specimen collection and submission for microbiology studies. Each specimen will be subject to the following;

1. JMH General Laboratory Protocols & Procedures
 - a) Specimen Organization, workflow & specimen retention-Standard protocol for specimen processing and specimen retention
 - b) Criteria for the Rejection of Specimens
 - c) Laboratory Specimen Collection Schedule
 - d) Processing Lab Specs for Non Patients
 - e) Laboratory Patient Identification & Specimen Labeling
2. JMH Microbiology Protocols & Procedures
 - a) Specimen Handling, Acceptability & Receipt Procedure
 - b) Specimen Rejection Procedure
 - c) Microbiology Test Request, Computer Charges & Form Requirements

II. Specimen Requirements:

A. All diagnostic information from the microbiology laboratory is contingent on the quality of specimen received. Consequences of a poorly collected and/or poorly transported specimen include failure to isolate a microorganism and recovery of contaminants or normal microbiota, which can lead to improper treatment of the patient. Often, direct specimen smears are utilized to determine the quality of the specimen, to provide rapid information for diagnosis and therapy, and to allow the physician to determine if additional, better-quality specimens should be collected.

B. General Consideration:

1. Safety

- a) **Follow universal precaution guidelines.**
- b) **Medical workers are to use appropriate barrier protection** (such as gloves and coat or gown) when collecting or handling specimens. If splashing may occur, protective eyewear, or face masks may be necessary.
- c) **Do not contaminate the external surface of the collection container and/or its accompanying paperwork.** Minimize direct handling of specimens in transit from the patient to the laboratory. **Use plastic bio-hazardous sealable bags** with a separate pouch for the laboratory requisition orders.
- d) **Centrifuged untreated specimens must loaded & unloaded in the bio-safety cabinet.**

- III. Reagents:**
1. Sterile Swab transport systems (Aerobic or Anaerobic)-Amies or Stuarts medium
 - a. BBL CultureSwab-LQ Stuart swab
 2. Calcium alginate swabs
 3. Dacron Swabs
 4. Nasopharyngeal-urethrogenital swabs
 5. Sterile screw cap cups
 6. Sterile petri dishes
 7. Sterile tubes (Vacutainer tubes- SPS yellow top w/o additives & chemistry green top with Na Hep)
 8. BACTEC Blood culture bottles
 9. 0.85% NaCl (Saline)
 10. Capped Sterile Syringe without needle
 11. Glass slides
 12. Povidone-Iodine Prep Pads

- IV. Procedure:**
1. **General guidelines for proper specimen collection** (See tables 1, 2, 3, 4, 5, 6, 7 & 8 for specific recommendations and considerations):
 - a. Collect specimens before administering antimicrobial agents when possible.
 - b. Collect specimen with as little contamination from indigenous microbiota as possible. Utilize appropriate sterile collection devices as indicated for each test. Use sterile equipment and aseptic technique to collect specimens. Ampules designed to keep swabs moist should be broken at the time of collection, when the swabs are inserted into the transport tube. Most specimens collected with a swab and transported dry are unacceptable. Do not use expired container or medium to transport specimen.
 - c. ***All specimens submitted to the laboratory must be properly identified by indicating the patient's name, encounter number, the date and time of collection on every specimen tube, slide or container submitted and source or site of specimen submitted.***
 - d. Collect an adequate amount of specimen.
 - e. Include "rule-out" request when ordering microbiology tests when appropriate. For example: Rule-out Meningitis on nasal specimens or MRSA. Record this information in the COMMENT FIELD IN STAR.
 - f. Identify the specimen source and/or specific site correctly so that proper culture media will be selected during processing in the laboratory.

- g. Collect specimens in sturdy, sterile, screw-cap, leak proof containers with lids that do not create an aerosol when opened. Collection containers are to be closed securely and precautions taken to prevent leaking of sample during transport. These specimens may be biohazard. Do not use petri dishes for specimen collection and transport.
 - h. Specimens obtained by a physician using needle aspiration should be transferred to a sterile tube or anaerobic transport vial prior to transport of the specimen to the laboratory. If there is little material in the syringe, the physician should draw a small amount of sterile nonbacteriostatic 0.85% NaCl or sterile broth through the syringe and transfer the specimen to a sterile tube. Alternatively, a small amount of sterile 0.85% NaCl or broth may be drawn into the syringe prior to removal of the needle. The physician should use a protective device while removing the needle to avoid injury and should cap the syringe with a sterile cap prior to transporting it to the laboratory. If a fairly large volume is collected (2ml or more), anaerobic bacteria survive for 24 h at room temperature.
2. **General guidelines for proper specimen transport** (See table 2 & 3 for specific recommendations):
- a. Transport all specimens to the laboratory promptly in sealed plastic bio hazardous bags with request form securely placed in a separate pouch or attached outside the bag.
 - b. Alternatives to prompt delivery.
 - 1) Refrigerate most specimens at 2 to 8°C. The following are exceptions.
 - a) For Blood cultures, hold at room temperature.
 - b) Specimens that may harbor temperature-sensitive organisms such a *Neisseria* species should be left at room temperature.
 - c) For anaerobic specimens, use anaerobic transport system or Port-a Cult Swabs.
 - d) Stool specimens:
 - (1) Inpatient specimens use sterile specimen cups for both stool cultures & Ova and parasitic studies.
 - (2) Outpatient specimens
 - (a) For bacterial culture, mix stool with a transport medium (such as Para-Pak C&S for culture & sensitivity or Cary Blair medium or buffered glycerol saline).
 - (b) For parasitology examination, mix stool with preservative such as Para-Pak Zn-PVA Fixative & 10% Buffered Neutral Formalin.
 - e) Hold CSF specimens at room temperature unless they are to be cultured for viral studies.

3. **Recommended guidelines for timing and storage of specimen received** (See table 2 for specific recommendations):

a. If no transport medium is used, specimens should be delivered and received in the laboratory within two hours after collection.

1) Since it is not always practical for many specimens to be inoculated upon receipt in the laboratory, refrigeration at 4° C to 6° C offers a safe and dependable method for storing certain clinical specimens until they can be processed. The length of time of specimen refrigeration varies with the type of specimen as indicated below.

a) Specimens that can be refrigerated without loss of pathogens:

- (1) Specimen Swabs used for wounds (except anaerobes), throat & streptococcus screens, and fecal specimen-2 to 3 hours.
- (2) Urine specimens-at least 24 hours (except TB specimens).
- (3) Viral specimens-over 24 hours or frozen at -70° C-see Quest reference lab

b) Specimens that cannot be refrigerated:

- (1) Fluid specimens such as blood, pleural, spinal fluids, joint & body fluids, etc...
- (2) Swab specimens for anaerobes, eye, ear, nasal, G.C., genital & cervical cultures.
- (3) Fecal specimens for O&P trophozoite studies

4. **Suggested Collection Instructions for Different Anatomic Sites:** (Not intended to replace JMH Nursing Procedures)

a. **Blood Cultures**

1) General considerations

a) Number & Timing

- (1) Acute sepsis-Collect two or three cultures from separately prepared sites prior to starting therapy.
- (2) Endocarditis
 - (a) Acute-Obtain three blood cultures with three separate venipunctures over 1 to 2 hours, and begin therapy.
 - (b) Subacute-Obtain three blood cultures on day 1 (15 min or more apart). If all are negative 24 hours later, obtain three more.
 - (c) Antimicrobial therapy 1 to 2 weeks before admission-Obtain two separate blood cultures on each of three successive days.

- b) Volume of blood
 - (1) The volume of blood is critical because the concentration of organisms in most cases of bacteremia is low, especially if the patient is on antimicrobial therapy. In infants and children, the concentration of organisms during bacteremia is higher than in adults, so less blood is required for culture. See table 2.
 - (a) Children: 1 to 5 ml of blood per venipuncture
 - (b) Adults: 10 to 30 ml of blood per venipuncture
 - c) Culture medium
 - (1) General consideration-Blood-to-medium ratios from 1:3 to 1:10 are recommended.
 - (2) Media-See table 2
- 2) Blood collection can be obtained by venipuncture of peripheral veins or arteries or from intravascular catheters.
- Observe universal precautions. Wear gloves.**
- a) Disinfect the venipuncture site and stoppers of culture bottles and collection tubes prior to blood collection. (Note: Do not use iodine to disinfect the stoppers of BACTEC bottles.
 - b) Swab concentrically, starting at the center with 1 to 2% tincture of iodine or 10% povidone-iodine solution. (If the patient is hypersensitive to iodine, prepare the skin by using a double application of 70% alcohol.)
 - c) Allow the disinfectant to dry. (Note: Do not palpate the vein after disinfecting the skin prior to inserting needle.)
 - d) Draw blood and deliver through a transfer set or a double-ended needle into a sterile collection bottle or tube. (A new needle or transfer set should be used for each venipuncture.)
 - e) After venipuncture and after inoculation of culture collection bottle or tube, wipe residual iodine from the skin with alcohol to prevent irritation of skin. Dispose of collection system in accordance with universal precautions.

b. Central nervous system (CNS) specimens

- 1) CSF-Suggested volumes are 1,2, and 2 ml for routine, fungal, and mycobacterial culture, respectively or as prescribed by JMH nursing procedures.
 - a) **Lumbar puncture** (Suggested Skin Prep & Specimen Collection)
 - (1) Clean the puncture site with antiseptic solution and alcohol before needle insertion to prevent introduction of infection.

- (2) Insert a needle with stylet at the L3-L4, L4-L5, or L5-S1 interspace. When the subarachnoid space is reached, remove the stylet and spinal fluid will appear in the needle hub.
 - (3) Slowly drain the CSF into the sterile leakproof tubes. Three tubes are generally required for microbiology, hematology, and chemistry testing. The second tube drawn will generally go to microbiology. (In traumatic taps, the CSF will often clear as the later tubes are collected.) Note: Always send the most turbid tube to microbiology.
- b) **Ommaya reservoir fluid**
- (1) Clean the Ommaya reservoir site with antiseptic solution and alcohol prior to removal of Ommaya fluid to prevent introduction of infection.
 - (2) Remove Ommaya fluid via the Ommaya reservoir unit, and place it in a sterile tube.
- c) Other CNS specimens
- (1) **Brain abscess**-Since 90% will grow anaerobic bacteria, the aspirated material from a lesion should be sent to the microbiology laboratory in an anaerobic transport system or transported without delay for immediate processing.
 - (2) **CNS biopsy** samples are obtained from the lesion at surgery, and is to be sent to the microbiology laboratory in an anaerobic transport system. Do not add formalin to microbiology specimens.
- c. **Gastrointestinal tract (includes the esophagus, stomach, duodenum, small intestine, and colon)**
- 1) **Fecal specimens** submitted primarily for the detection of *Campylobacter*, *Shigella*, *Salmonella* species, *Clostridium difficile* toxin A antigen screens and in certain cases to detect *Yersinia*, *Vibrio*, *Aeromonas* species and enterotoxigenic *Escherichia coli*.
 - a) General considerations
 - (1) Keep stool specimens cool; do not incubate them.
 - (2) If a stool specimen cannot be planted within 1 hour of collection, it should be mixed with transport medium.
 - (3) Do not use toilet paper to collect stool.
 - b) Have patient obtain stool specimen by one of the following methods.
 - (1) Pass stool directly into a sterile, wide-mouth, leakproof container with a tight-fitting lid.
 - (2) Pass stool into a clean, dry bedpan, and transfer stool into a sterile leakproof container with a tight-fitting lid.
 - 2) **Rectal swabs**-Primarily submitted for detection of *Neisseria gonorrhoeae*, *Shigella* species, herpes simplex virus (HSV), and anal carriage of *Streptococcus pyogenes*.

- a) Pass the tip of a sterile swab approximately 1 in. beyond the anal sphincter. Carefully rotate the swab to sample the anal crypts, and withdraw the swab. Send the swab in a swab transport system.
- 3) **Gastric lavage**-Primarily submitted for the detection of *Mycobacterium tuberculosis* in patients unable to produce quality sputum.
 - a) Pass a well-lubricated tube orally or nasally through to the stomach of the patient, and perform the lavage. Before removing the tube, release the suction and clamp to prevent mucosal trauma and/or aspiration.
 - 4) **Duodenal aspiration**-Primarily submitted for the detection of *Giardia* species and larvae of *Strongyloides stercoralis* and *Ascaris lumbricoides*.
 - a) Pass a tube orally through to the duodenum of the patient.
 - b) To aspirate a sample for giardiasis, the tube should be at least in the third portion of the duodenum.
 - 5) **Gastric biopsies and washings**-patient should fast prior to each of the following procedures.
 - a) **Esophageal specimens** are primarily used to detect *Candida* species, cytomegalovirus (CMV), and HSV infections. **Stomach and duodenum specimens** are used for detection of *Helicobacter pylori*. **Duodenum specimens** can also be used for detection of *Giardia* species and the larvae of *S. stercoralis* and *A. lumbricoides*.
 - (1) Pass an endoscope orally.
 - (2) Obtain specimens through a channel in the endoscope by using one of the following procedures.
 - (a) Using biopsy forceps, obtain samples from the esophagus, stomach, or duodenum.
 - (b) Using a sheathed brush, brush suspicious areas several times to obtain adequate cellular material.
 - (c) Perform a wash by injecting approximately 25 to 30 ml of sterile nonbacteriostatic isotonic 0.85% NaCl through the biopsy channel onto the lesion. Collect the specimen by aspirating the fluid through the scope into a sterile trap, which is connected to the suction tubing. Note: If a gastric ulcer is seen, obtain biopsy samples from the base, the surrounding gastric mucosa, and each of the four quadrants of the margin.
 - b) **Rectal biopsy**-Primarily submitted for detection of *Entamoeba histolytica*, *Balantidium coli*, and HSV. If lesions are not evident, biopsy the posterior rectal mucosa below the peritoneal reflection (within 7 to 10 cm of the anal verge).
 - c) **Small bowel biopsy**-Primarily submitted for detection of *Giardia*, *Cryptosporidium*, and *Microsporidium* species.
 - d) **Sigmoidoscopy**-Useful in detection of *E. histolytica* and *Mycobacterium* species and the diagnosis of pseudomembranous colitis associated with *C. difficile* and possibly *Staphylococcus aureus*.

- (1) Perform flexible or rigid sigmoidoscopy.
- (2) Obtain endoscopic pinch biopsy samples of any lesions seen. Additionally, aspirate liquid from the inflamed bowel with a pipette passed through the sigmoidoscope. Transport specimens in a sterile screw-cap container. If biopsy samples are small, add a small amount of sterile nonbacteriostatic 0.85% NaCl to prevent the specimen from drying.

d. **Genital tract specimens**

- 1) **Female**-Primarily submitted for detection of sexually transmitted pathogens (such as *N. gonorrhoeae*, *Chlamydia trachomatis*, lymphogranuloma venereum, HSV, human papillomavirus (HPV), trichomonads, *Haemophilus ducreyi*, group B streptococci, and *Candida* infections). If infection is not caused by any of these pathogens, anaerobic bacteria may be involved.
 - a) **Amniotic fluid**-Aspirate fluid by catheter, at cesarean section, or at amniocentesis.
 - b) **Bartholin gland**-Decontaminate the skin with povidone-iodine, and aspirate material from the duct(s).
 - c) **Cervix**
 - (1) Do not use lubricant during procedure
 - (2) Wipe the cervix clean of vaginal secretion and mucus.
 - (3) Rotate a sterile swab, and obtain exudates from the endocervical glands.
 - (4) If no exudates is seen, insert a sterile swab into the endocervical canal, and rotate the swab.
 - d) **Endometrium specimens** are collected by transcervical aspiration through a telescopic catheter.
 - e) **Fallopian tube specimens** are obtained by aspiration or by swab during surgery. Bronchoscopy cytology brushes may be used if exudate is not expressed.
 - f) **Rectal swabs** are used primarily to detect *N. gonorrhoeae*, *Shigella* species, HSV, and anal carriage of *S. pyogenes*.
 - g) **Urethra**
 - (1) Collect specimens 1 hour or more after patient has urinated.
 - (2) Stimulate discharge by gently massaging the urethra against the pubic symphysis through the vagina.
 - (3) Collect the discharge with a sterile swab.
 - (4) If discharge cannot be obtained, wash external urethra with betadine soap and rinse with water. Insert a urethrothra swab 2 to 4 cm into the endourethra, gently rotate the swab, and leave it in place for 1 to 2 seconds. Withdraw the swab, and submit it in the appropriate transport system for culture.
 - h) **Vagina specimens** are useful in the detection of group A streptococci in children.
 - (1) Use a speculum without lubricant and collect secretions from the mucosa high in the vaginal canal with a sterile pipette or swab.

i) **Vulva**

- (1) Clean the surface of the lesion with 0.85% NaCl. If there is a crust on the lesion, remove it.
- (2) Scrape the lesion until serous fluid emerges.
- (3) Wipe away fluid and debris with sterile gauze.
- (4) Press the base of lesion until clear fluid is expressed.
- (5) Aspirate vesicular fluid with a 26- to 27-gauge needle.
OR
- (6) Touch a slide to the fluid, and cover the fluid on the slide with a coverslip (for *Treponema pallidum* detection). OR
- (7) Unroof the vesicle, and collect fluid with a sterile swab (for HSV detection). OR
- (8) Scrape the base of an open vesicle with a sterile scalpel blade, and then rub the base vigorously with a sterile swab (for HSV and *H. ducreyi* detection).

2) **Male**

- a) **Anal Swab** is primarily submitted for detection of *N.gonorrhoeae*, *Shigella* species, HSV, and anal carriage of *S. pyogenes*.
 - (1) Pass the tip of a sterile swab approximately 1 in. beyond the anal sphincter. Carefully rotate the swab to sample the anal crypts, and withdraw it. Send the swab in a swab transport, viral transport (for HSV), or *N. gonorrhoeae* transport system.
- b) **Epididymis specimen** is primarily used to diagnose nonspecific bacterial epididymitis (most commonly due to members of the family *Enterobacteriaceae* or pseudomonads and generally occurs in men over 35 years), sexually transmitted epididymitis (most commonly due to *C. trachomatis* and *N. gonorrhoeae*) and *M. tuberculosis* infections (generally occur after involvement of the prostate or seminal vesicles).
 - (1) Use a needle and syringe to aspirate material from the epididymis.
- c) **Penile lesion specimens** are primarily used to detect sexually transmitted pathogens such as *N. gonorrhoeae*, *C. trachomatis*, lymphogranuloma venereum, HSV, *T. pallidum*, and *H. ducreyi*.
 - (1) Clean the surface of the lesion with 0.85% NaCl. If there is a crust on the lesion, remove it.
 - (2) Scrape the lesion until serous fluid emerges.
 - (3) Wipe away fluid and debris with sterile gauze.
 - (4) Press the base of lesion until clear fluid is expressed.
 - (5) Aspirate vesicular fluid with a 26- to 27-gauge needle.
 - (6) Touch a slide to the fluid, and cover the fluid on the slide with a coverslip (for *T. pallidum* detection). OR
 - (7) Unroof the vesicle, and collect fluid with a sterile swab (for HSV detection). OR

- (8) Scrape the base of an open vesicle with a sterile scapel blade, and rub the base vigorously with a sterile swab (for HSV and *H. ducreyi* detection).
- d) **Prostatic massage specimens** are primarily used to diagnose acute or chronic prostatitis. For both diseases, gram-negative enteric organisms are the most frequently isolated pathogens. *N. gonorrhoeae* is found infrequently.
 - (1) Perform a digital massage through the rectum.
 - (2) Collect the specimen in a sterile tube or on a sterile swab.
- e) **Urethra specimens** are primarily used to detect *N. gonorrhoeae* and *C. trachomatis*.
 - (1) Collect specimens at least 2 hours after the patient has urinated.
 - (2) Insert a thin urethrogenital swab 2 to 4 cm into the endourethra, gently rotate it, leave it in place for 1 to 2 seconds, and withdraw it.

e. **Ocular specimens**

- 1) General considerations
 - a) Obtain viral and chlamydial samples before topical anesthetics are instilled.
 - b) Obtain samples for chlamydial cultures with calcium alginate swabs and for viral cultures with Dacron swabs or cotton swabs with nonwood shafts.
 - c) Send prepared smears and inoculated media to the laboratory immediately.
- 2) **Conjunctival scrapings**
 - a) One or 2 drops of topical anesthetic are generally instilled.
 - b) Scrape the lower tarsal conjunctiva with a sterilized kimura spatula.
 - c) Inoculate the appropriate media directly.
 - d) Prepare smears by applying the scraping in a circular manner to a clean glass slide or by compressing material between two glass slides and pulling the slides apart.
 - e) Alternatively, use a calcium alginate swab or a cotton-tipped applicator to swab the inferior tarsal conjunctiva (inside surface of eyelid) and the fornix of the eye.
- 3) **Corneal scrapings**
 - a) Obtain conjunctival samples prior to corneal scrapings.
 - b) One or 2 drops of topical anesthetic are generally instilled.
 - c) Using short, firm strokes in one direction, scrape multiple areas of ulceration and suppuration with a sterilized kimura spatula. (Keep the eyelid open, and be careful not to touch the eyelashes.)

- V. **References:** 1. Isenburg, H.D., Clinical Microbiology Procedures Handbook, Vol 1, 1992; Section Aerobic Bacteriology-Specimen Collection and Transport, pgs 1.1.1 – 1.1.29.

Table 1 Transport systems for aerobic specimens

| Systems | Comments |
|---|--|
| Swab Transport System (Aerobic BBL CultureSwab Set-ups such as Cary-Blair transport medium, Amies charcoal medium, LQ Stuart), Cultorettes and Anaerobic Transport systems) | Sterile, disposable culture collection & transport system consisting of plastic tube containing rayon-tipped swabs & transport medium to prevent drying of bacteria & maintain pH. (Note: Always crush ampoule-w/protective sleeve over ampoule-after specimen collection. Cary-Blair transport medium allows recovery of <i>Salmonella</i> & <i>Shigella</i> , <i>Vibrio cholerae</i> , & <i>Campylobacter</i> from fecal spec. Amies charcoal medium is good for <i>N. gonorrhoeae</i> & other fastidious organisms. <i>Yersinia pestis</i> survives for at least 75 days. |
| Calcium alginate swabs | Can be toxic for some strains of <i>N. gonorrhoeae</i> , HSV, & <i>Ureaplasma urealyticum</i> & may be toxic for some cell cultures. Useful for collection of <i>Chlamydia</i> cultures. |
| Dacron/Polyester tipped swabs | Useful in collection of viral & group A streptococcus specimens |
| Nasopharyngeal-urethrogenital swabs | Flexible wire shafts & small tips provide easier specimen collection, esp. for collection of nasopharyngeal specimens, <i>Bordetella pertussis</i> , & male urethral specimens for <i>N. gonorrhoeae</i> . |
| Sterile screw cap cups | Useful for collection of urine, sputum, stool, bronchoalveolar lavage, & biopsy specimens. If biopsy specimen is small, add small amount of sterile nonbacteriostatic 0.85% NaCl in to cup. Never place microbiology biopsy specimens in formalin or wrap in gauze. |
| Sterile tubes (screw-cap glass or plastic tubes, sterile Vacutainer tubes w & w/o additives) | Useful for collection of sterile fluids, bronchoalveolar lavage, drainage, or brush specimens. Yellow top & Green top w Na Hep Vacutainer tubes. |
| Reference Lab Transport Systems | A. Quest Lab 1. For <i>Mycoplasma/Ureaplasma</i> 2. Cultures use <i>Mycoplasma pneumonia</i> Transport Medium; for genital mycoplasma use medium labeled "M4"; 3. For ova & parasitic studies use Para-Pak O&P kits; 4. For viral cultures use a universal transport medium; 5. For Blood/Body fluid Fungal studies use Chem Green top tubes with Na Hep, and for other specimens see Quest reference instructions B. CT State Lab-Send Out Containers (TC, RC, PP, & WC) |

Table 2 Specimen transport guide

| Source & type of specimen | Transport method |
|---|---|
| Blood | <p>Blood transport system (BACTEC btles) or sterile yellow top tube with SPS. If <i>Neisseria</i> spp. Are suspected, use transport system w/o SPS: Transport Time <4 hrs; Room Temperature</p> <ol style="list-style-type: none"> Routine Cultures-2 separate venipunctures drawn at least half hour apart/Max of 3 set per day <ul style="list-style-type: none"> BACTEC STD/10 Aerobic/F Culture vial (Optimal 8-10 ml/range 3 to 10 ml) BACTEC STD Anaerobic/F Culture vial (Optimal 5-7 ml/range 3-7 ml) BACTEC PEDS PLUS/F Culture vial-Difficult/small volumes (Optimal 1-3 ml/range 3 to 10 ml) Stat Cultures-2 separate venipunctures (different sites) drawn at the same time intervals Special Request/Per Doctor's Orders <ol style="list-style-type: none"> Adult & Adolescent Patient <ol style="list-style-type: none"> Sever septicemia, meningitis, arthritis, osteomyelitis, pneumonia-2 sets prior to therapy Sub acute Bacterial Endocarditis (S.B.E.)-3 Sets w/in 24 hrs-spaced 15 min or more apart. If all negative 24 h later, obtain 3 more Acute Bacterial Endocarditis (A.B.E.)-3 Sets w/in 1 to 2 hrs before therapy Low-Grade Intravascular Infections-3 Sets w/in 24 hrs Antimicrobial therapy 1 to 2 weeks before admission-2 separate cultures on each of 3 successive days Fever of Unknown Origin-2 separate culture at least 1 h apart. If negative, then 24 to 36 h later, obtain 2 more cultures 1 h apart. Young Children: (2 Set-half hr apart) <ol style="list-style-type: none"> Infant-2yrs: 1 to 3 ml 2) 2 to 4 yrs: 2 to 4 ml 3) 5 to 6 yrs: 4 to 6 ml 7 to 10 yrs: 6 to 10 ml 5) 10 to adult: 16 to 20 ml |
| Central Nervus System CSF Ommaya fluid Brain abscess CNS biopsy | <p>Transport Time-Stat; Room Temperature</p> <p>Sterile Screw-cap tube</p> <p>Same as above</p> <p>Anaerobic transport system (swabs or syringe)</p> <p>Same as above. If specimen is small, send in sterile cup w/small amount of nonbacterostatis 0.85% NaCl (Saline). <i>Never place in formalin.</i></p> |
| Gastrointestinal system Feces Rectal swab Gastric lavage or washings Duodenal aspirate Rectal biopsy Sigmoidoscopy spec | <p>Transport Time: Deliver to lab w/in 24 hrs/For Yersinia, deliver to lab w/in 4 hrs of collection; Room Temperature or refrigerate if delay in transport occurs</p> <p>Sterile screw-cap or tube w/Cary Blair medium or buffered glycerol saline (R/O enteric pathogens)</p> <p>Routine cultures accept One Stool or Rectal Swab spec per patient per Day; For Carrier detection-3 specs over a 3 day period-one per day</p> <p>Swab transport system (For Pinworm, use pinworm collection kit)</p> <p>Sterile screw-cap cup or sputum trap</p> <p>Same as above</p> <p>Sterile screw-cap cup or tube. If specimen is small, send in sterile cup with small amount of nonbacteriostatic 0.85% NaCl (Saline). <i>Never place in formalin.</i></p> <p>Same as above</p> |
| Eye Conjunctiva scrapings Corneal scrapings Intraocular fluid Surface/Drainage | <p>Transport Time-ASAP; Room Temperature</p> <p>Send prepared smear and directly inoculated media</p> <p>Same as above</p> <p>Same as above, Anaerobic transport system, or capped syringe without needle with air expelled.</p> <p>Swab transport system-Calcium alginate swab recommended</p> |
| Genital tract, female Amniotic fluid Fallopian tube Bartholin fluid Culdocentesis Cervical Urethral Vaginal Endometrial Vulval | <p>Transport Time-Routine; Room Temperature</p> <p>Anaerobic transport system with 1-2 ml of sample</p> <p>Same as above</p> <p>Same as above</p> <p>Same as above</p> <p>Swab transport system, viral or chlamydial transport, or <i>N. gonorrhoeae</i> transport system</p> <p>Same as above</p> <p>Same as above</p> <p>Sterile screw-cap or tube or anaerobic transport system</p> <p>Capped syringe without needle; swab transport system, viral or chlamydial transport, or <i>N. gonorrhoeae</i> transport system</p> |
| Ear Canal/Drainage | <p>Transport Time-Routine; Room Temperature</p> <p>Swab transport system</p> |
| Diphtheria Culture Nasopharynx | <p>Transport Time-Routine; Room Temperature</p> <p>Use Conn State Lab <i>B.pertussis</i> "WC" Kit-Sent CT Dept of Health</p> |
| Pinworm exam | <p>Transport Time-Routine; Room Temperature</p> <p>Pinworm Collection Kit-Early AM spec prior to using lavatory</p> |
| Tick ID | <p>Transport Time-Routine; Room Temperature</p> <p>Screw top lid container</p> |

Table 2 Specimen transport guide-Continued

| | |
|---|--|
| Clostridium difficile EIA Toxin A Scn | Transport Time-Routine; Temp-Refrigerate Screw Cap Container |
| Specimens for <i>N. gonorrhoeae</i> Anal, cervical, urethral, vaginal | Transport Time-Submit within 6 hrs; Room Temperature Swab transport system |
| Lower respiratory tract Lung biopsy Expectorated sputum Induced sputum Tracheal or endotracheal aspirate Bronchoalveolar lavage fluid Bronchial washings Transbronchial biopsy Bronchial brush Transtracheal aspirate Lung aspirate | Transport Time-Routine: Room Temperature Sterile screw-cap cup; if specimen is small, place it in a small amount of nonbacteriostatic 0.85% NaCl (Saline). <i>Never place in formalin</i> Sterile screw-cap cup (Saliva is unacceptable) Sterile screw-cap cup Sputum trap or sterile screw-cap cup or tube Same as above Same as above Sterile screw-cap tube with 1-2 ml of nonbacteriostatic 0.85% NaCl (Saline) Same as above Anaerobic transport system or sterile screw-cap cup or tube Same as above |
| Upper respiratory tract Throat Nasal Oral culture Nasopharyngeal swab Tympanocentesis fluid Sinus aspirate Nasopharyngeal suction Nasal washings | Transport Time-Routine; Room Temperature Swab transport or virus transport system-for viral culture studies Same as above Same as above Same as above Anaerobic transport system or capped syringe without needle Same as above Sterile screw-cap cup or viral transport system Same as above |
| Sterile body fluids (excluding CSF, urine, blood) Pleural, peritoneal, ascites, joint, & synovial fluids, etc.. | Transport Time-ASAP; Room Temperature (Abdominal, Amniotic, Ascitic, Bile, Bone Marrow, Chest, Dialysate, Pancreatic, Paracentesis, Pericardial, Peritoneal Dialysate, Peritoneal, Plural & Thoracentesis fluid) Sterile screw-cap container, capped syringe without needle, or anaerobic transport system |
| Subcutaneous tissue & skin Ulcers or nodules, superficial wound (bacterial) Exudate Biopsy Burn specimens Superficial fungal lesion material | Transport Time-Routine; Room Temperature Capped syringe without needle Sterile screw-cap cup (If specimen is small, add a small amount of sterile nonbacteriostatic 0.85% NaCl (Saline) to prevent drying. Sterile screw-cap container Same as above |
| Deep wounds, aspirate, tissues Site wound Deep wounds or abscesses Soft tissue aspirates Bone Punch skin biopsy | Transport Time-Routine/ASAP; Room Temperature Swab transport system-Aerobic & Anaerobic or Sterile screw-cap container Capped syringe without needle Sterile screw-cap container (If specimen is small, add a small amount of sterile nonbacteriostatic 0.85% NaCl (Saline) to prevent drying) Same as above |
| Ova & Parasite Studies Fresh stool-esp bloody, mucoid or watery samples | Transport Time-No Time Limit; Room Temperature Sterile Screw Capped Container or Para-Pak 10% Buffered Formalin & Zn-PVA Fixative Kit Recommend collecting a minimum of 3 spec over 3 consecutive days (one each day); To R/O amoebiasis, 6 specimens are recommended. |

Table 2 Specimen transport guide-Continued

| | |
|--|--|
| Urine Clean catch Ileal conduit Straight catheter | Transport Time-Max/Within One hour at Room Temperature ;If > 1 hour, refrigerate; No. of spec- one spec per day Sterile screw-cap cup or tube or urine collection tube Same as above |
| Suprapubic aspirate Bladder washout Bilateral ureteral catheterization | Transport Time-ASAP; Room Temperature Anaerobic transport system or capped syringe without needle Sterile screw-cap or tube (Be careful to label specimens with correct times & sites) Same as above |

Table 3 Specimen Requirements for Mycobacterial Cultures & Acid-Fast Stains

| Specimen type | Specimen Requirements | Special Instructions | Unacceptable Specimens |
|--|--|--|--|
| Abscess contents, Aspirated fluid | As much as possible in syringe with Luer tip cap | Cleanse skin w/alcohol before aspirating sample. Collect spec on swab, & place in transport medium or if volume is sufficient for needle & syringe, | Dry swab |
| Blood | 10-ml SPS (yellow top) Blood collection tube | Disinfect site as for routine blood culture. Mix tube contents immediately after collection. SPS is preferred anticoagulant. Heparinized blood is also ok | Blood collected in EDTA. Coagulated Blood |
| Body fluids (pleural, pericardial, peritoneal, etc.) | As much as possible (10-15 ml min) in sterile container or syringe w Luer tip cap. Collect bloody specs into SPS blood collection yellow top tubes | Disinfect site w alcohol if collected by needle & syringe. Use conical polypropylene bottles (200 ml) for centrifugation of large volumes. Since many of these fluids may contain fibrinogen, it may be necessary to add anticoagulant to collection containers. | |
| Bone | Bone in sterile container With out fixative or preservative | | Specimen submitted in formalin |
| Bone marrow | As much as possible in SPS- yellow top Blood collection tube | Collect aseptically. Mix SPS tube contents immediately following collection | |
| Bronchialveolar Lavage or Bronchial washings | >/=5 ml in sterile container | Avoid contaminating bronchoscope with tap water. Saprophytic mycobacteria may produce false-positive or smear results | |
| Bronchia brushing | Sterile container | | |
| CSF | >/=2 ml in sterile container | Use maximum volume attainable | |
| Gastric lavage fluid | >/=5-10 ml in sterile container. Collect in the morning soon after patient awakens. | Collect fasting early-morning spec on 3 consecutive days. Use sterile saline. | Specimen that has not been neutralized |
| Lymph node | Node or portion in sterile container without fixative or preservative | Collect aseptically, & avoid indigenous microbiota. Select caseous protein if available. Do not immerse in saline or other fluid or wrap in gauze. Freezing decreases yield. | Specimen submitted in formalin |
| Skin lesion material | Submit biopsy spec in sterile container without fixative or preservative. Submit aspirate in syringe with Luer tip cap. | Swabs in transport medium (Amies or Stuarts) are acceptable only if biopsy sample or aspirate is not obtainable. For cutaneous ulcer, collect biopsy sample from periphery of lesion. If infection was acquired in Africa, Australia, Mexico, South America, Indonesia, New Guinea, or Malaysia, note on request, because <i>M.</i> <i>ulcerans</i> may require prolonged incubation for primary isolation. | Dry swab |
| Smear on slides | Smear spec over 1.5-by 1.5-cm area of clear slide | Heat fix smears. Transport in slide container taped closed & labeled BIOHAZARD. | |

Table 3 Specimen Requirements for Mycobacterial Isolation & Acid-Fast Stains-Continued

| Specimen type | Specimen requirements | Special Instructions | Unacceptable specimens |
|------------------------|--|---|---|
| Sputum | 5-10 ml in sterile, wax-free disposable container. Collect an early-morning spec from deep, productive cough on at least 3 consecutive days. Follow-up of patient on therapy, collect at weekly intervals beginning 3 weeks after initiation of therapy. | For expectorated sputum, instruct patient on how to produce sputum spec as distinct from saliva or nasopharyngeal discharge. Have patient rinse mouth with water before collecting sputum to minimize contamination. For induced sputum, use sterile hypertonic saline. Avoid sputum contamination with nebulizer reservoir water. Indicate on request if spec is induced sputum. | 24-h pooled spec; saliva |
| Stool | >= 1 g in sterile, wax-free disposable container | Collect spec directly into container, or transfer from bedpan or plastic wrap stretched over toilet bowl. Was from container may produce false-positive smear | Specs submitted in formalin |
| Tissue biopsy | 1 g of tissue, if possible, in sterile container without fixative or preservative. | Collect aseptically, & avoid indigenous microbiota. Select caseous portion if available. Do not immerse in saline or other fluid or wrap in gauze. Freezing decreases yield. | Specs submitted in formalin |
| Transtracheal aspirate | As much as possible in syringe with Luer tip cap or other sterile container | | |
| Urine | As much as possible (min 40 ml) of 1 st morning spec obtained by catheterization or of midstream clean catch in sterile container. For suprapubic tap, as much as possible in syringe with Luer tip cap or other sterile container | Collect 1 st morning spec on 3 consecutive days. Accept only 1 spec per day | 24-h pooled specs Urine from catheter bag; Specs of <40 ml unless larger volume is not obtainable |
| Wound material | See biopsy or aspirate. | Swabs are acceptable only if biopsy or aspirate is not obtainable. If used, they must be placed in transport medium (Amies or Stuarts). Negative results are not reliable. | Dry swab |

Table 4 Collection consideration for respiratory specimens

| Culture | Vol (ml) | Comments |
|--------------------------|----------|--|
| Bacteria | NA | Contact laboratory if <i>Legionella</i> suspected. Submit sputum only; saliva is unacceptable. |
| Fungi | 3-5 | Collect three early-morning fresh specimens resulting from deep cough or sputum induction. Lung biopsy specimens or lung aspirates are also appropriate. |
| Anaerobes | 1 | Sinus aspirate, tympanocentesis fluid, transtracheal aspirate, & lung aspirates or biopsy specimens are appropriate. |
| Mycobacteria | 5-10 | Collect three early-morning fresh specimens resulting from deep cough or sputum induction. Lung biopsy specimens or lung aspirates are also appropriate. |
| <i>Pneumocystis spp.</i> | 2 | Use induced sputum, bronchoalveolar lavage fluid, or lung biopsy specimen. |
| Parasites | 3-5 | Can be examined for amoebae, helminth eggs (<i>Paragonimus westermani</i>), hooklets of <i>Echinococcus spp.</i> , larvae of hookworm, and <i>Ascaris & Strongyloides spp.</i> |

Table 5 Collection considerations for sterile body fluids

| Culture | Vol (ml) | Comments |
|--------------|----------|--|
| Bacteria | 1-5 | If gonococcal arthritis is suspected, notify laboratory to add modified Thayer-Martin plate. |
| Fungi | >10 | Boldd for <i>Histoplasma capsulatum</i> (AIDS), <i>Cryptococcus spp.</i> , <i>Candida albicans</i> , and <i>Candida tropicalis</i> |
| Anaerobes | 1-5 | Use anaerobic transport system |
| Mycobacteria | >10 | |

Table 6 Collection consideration for subcutaneous tissue & skin specimens

| Culture | Comments |
|--------------|--|
| Bacteria | Syringe aspirates or biopsy specimens are preferable to swab specimens. |
| Anaerobes | Uncommon in burn, ulcer, nodules, or superficial skin infections; useful following bites and trauma |
| Fungi | Useful in diagnosing dermatophytes, yeast, filamentous fungi, and dimorphic fungi |
| Mycobacteria | Useful in diagnosing <i>Mycobacterium marinum</i> , <i>Mycobacterium fortuitum</i> , and <i>Mycobacterium chelonae</i> |
| Virus | Useful in diagnosing HSV and varicella-zoster virus |

Table 7 Collection considerations for deep wound, aspirate, and tissue specimens

| Culture | Comments |
|-----------|--|
| Bacteria | Biopsy specimens or aspirates are better than swab specimens |
| Anaerobes | Useful in diagnosing actinomycosis; send in anaerobic transport system |
| Fungi | Useful in diagnosing <i>Pseudallescheria boydii</i> , <i>Bipolaris spp.</i> , <i>Exophiala spp.</i> , and <i>Fusarium spp.</i> |

Table 8 Collection consideration for urine specimens

| Culture | Vol (ml) | Comments |
|--------------|-----------------|---|
| Bacteria | 0.5-1 | Do not collect 24-h specimen. After proper cleaning of patient, use first morning midstream void. |
| Fungi | >20 | Do not collect 24-h specimen. First morning void is recommended. |
| Mycobacteria | >20 | Do not collect 24-h specimen. First morning three consecutive voided urine specimens are recommended. |
| Anaerobes | 1 | Use suprapubic aspirate. Send in anaerobic transport system. |
| Virus | 10-50 | Do not collect 24-h specimen. First morning void is recommended. Useful for adenovirus, mumps, and CMV detection. Send on ice, and transport to laboratory immediately. |
| Parasites | 24-h collection | Useful for detecting <i>Shistosoma haematobium</i> eggs, <i>Tricomonas vaginalis</i> trophozoites in males, and <i>Onchocerca volvulus</i> microfilariae. |