

App. R - Microbiology Specimen Collection-Table 1

TABLE I Specimen Collection: Routine Microscopy or Bacterial Culture

Specimen	Patient Preparation	Collection technique	Optimal specimen and volume	Collection container	Comments	Optimal Transport time
ANAEROBIC CULTURE	See specific site listed below.	Avoid contamination with normal flora. Obtain aspirate by needle and syringe.	Tissue or 1 mL or more aspirated from a closed space	Special anaerobic transport tube or syringe. Lab will not accept needle on syringe; replace needle with sterile Luer tip cap. Dry swabs will not be cultured.	Send to lab without delay. Aspirates and biopsies are preferable to swabs. Anaerobes are part of the normal flora of the mouth, intestinal and vaginal tract. Therefore, culture results from these sites must be interpreted with care.	*2 hours
BIOPSY	see Tissue					
BLOOD						
-Peripheral – UW patients	Skin decontaminated with iodine followed by 70% alcohol. Allow each to act 1 minute.	Sterile venipuncture. Avoid drawing specimen through catheter or cannulas. Decontaminate bottle tops with 70% alcohol only.	Adults & older children: 16-20 mL Infants: 0.5-3 mL	One set consists of one each BACTEC Plus aerobic (8-10 mL) and BACTEC Lytic anaerobic (8-10 mL) bottles for adults. One BACTEC Pedsplus bottle (0.5-3 mL) for infants & small children.	Initial Evaluation: Collect 2-sets from different sites before administration of antibiotics. For FUO an additional set may be submitted one or more hours later for a total of 3 sets within a 24-hour period. If negative, repeat the 3-set collection.	*2 hours blood culture bottles
-Peripheral – Outside patients				20 mL in yellow top tube		
-Bone Marrow	Same for blood culture above	Sterile percutaneous aspiration	1 mL or more	One BACTEC Pedsplus bottle (0.5 – 3 mL)		*1 hour

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BODY FLUIDS OR SECRETIONS						
(Other than blood, urine, or CSF): -Joint fluid -Hematomas -Pericardial fluid -Peritoneal fluid	Skin decontamination	Aspirate into syringe (exclude air) For cell count collect in lavender top (EDTA) vacutainer tube	1 mL or more	Peritoneal or Ascites fluid: direct inoculation into blood culture bottles is preferred. See Blood above. Other fluids: Syringe with needle replaced by sterile Luer tip (available from Materials Management), or sterile container.	Send to lab without delay	*2 hours (1 hour, if cell count ordered)
-Bile	Surgery	Aspiration with syringe during surgery or from post-op drainage site, or via nasogastric tube from duodenum	Several mL	Sterile container or syringe with needle replaced by a sterile Luer tip cap	Discard the first few mL from drainage which often contains contaminants	*2 hours
-Breast milk	Skin decontamination	Gentle manual expression	Several mL	Sterile container		*2 hours
BRONCHIAL WASHING See Respiratory tract						
CATHETERS (TIPS)						
-Foley catheter			Not accepted		Foley tips or catheters are unsuitable for routine bacterial cultures.	NA
-Vascular cannulae, -Central venous pressure lines, -Umbilical or intravenous catheters	Skin decontamination	Use a sterile scissor to cut off tip and/or 5-7 cm of intra- cutaneous segment or if <7 cm, submit entire catheter		Sterile container	Semi-quantitative culture of intracutaneous segments aids diagnosis of sepsis.	*1 hour

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CENTRAL NERVOUS SYSTEM						
-Brain biopsy	Surgical	Sample infected site rapidly.	Tissue	Special anaerobic transport tube or other sterile container transported immediately	Send to lab without delay. Do not add saline or other fluid to specimen	*1 hour
-Cerebrospinal fluid	Skin decontamination	Sterile lumbar puncture. Ventricular or suboccipital tap	1-5 mL	Sterile, clean screw capped plastic or glass tubes	All CSF specimens are STAT. Transport to lab immediately at room temp. (DO NOT put on wet ice).	*1 hour
-Meningomyelocele	Same as CSF	Sterile aspiration through skin	1-5 mL	Syringe with needle replaced by sterile Luer tip cap, or screw capped sterile tubes	Often contaminated or infected with skin flora	*1 hour
-Shunt fluid	Skin and catheter decontamination	Sterile aspiration through shunt	1-5 mL	Same	As above	*1 hour
DIALYSIS FLUID						
	Skin decontamination	Collect outflow fluid	100 mL	Sterile container or syringe with needle replaced by sterile Luer tip cap		*2 hours
EAR						
-Internal	Cleanse external canal with mild antiseptic	For intact ear drum, aspirate with syringe. For ruptured ear drum, collect on flexible-shaft swab using auditory speculum.	Swab or aspirated fluid	Sterile container or syringe with needle replaced by sterile Luer tip cap; swab in Amies transport medium		*1 hour

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-External	Cleanse external canal with mild detergent	Obtain specimen from active margin, preferably including fresh secretion from deeper areas	Swab or aspirated fluid	Sterile container; swab in Amies transport media	Dry swabs will be rejected.	*2 hours
EYES						
-Internal	Surgical specimen	Surgical technique	As much as possible	Sterile container	Carefully label left and right eye. Speed in transport and care in handling are very important. <u>Send to lab immediately.</u>	*1 hour
-External	Cleanse skin around eye with mild anti-septic Remove ointment, etc. with sterile cotton and saline.	Swabbing: pass moistened swab twice over lower conjunctiva. Avoid eyelid border and lashes. Scrapings: use local anesthetic and platinum spatula.	Moistened swabs Contact lab supervisor for direct inoculation of media	Sterile tubes with a small amount of tryptic soy broth available in Micro Lab.	Scrapings should be done by ophthalmologist. For diagnosis of viral or chlamydial infections and for cytology; conjunctival or corneal scrapings are necessary. (For <i>Acanthamoebae</i> cultures: see Parasitology Table)	*1 hour
FLUIDS	See Body Fluids					
GASTROINTESTINAL TRACT						
-Duodenal aspirate		Aspiration through tube	Several mL	Sterile container		*2 hours

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-Feces	None	Directly into any clean container	UW patients - several mL. Deliver to lab daily for 3 consecutive days. Outside patients – clean container or Cary-Blair transport medium.	Waxed cardboard or urine container with tight-fitting lid	Avoid contamination with urine, soap, etc. Send to lab without delay. Do not refrigerate. Stools cultures are not performed on patients hospitalized more than 3 days.	*2 hours
-Gastric aspirate	Nothing by mouth (NPO) for 10 hours before test	Through tube. To be done by physician	3-5 mL	Sterile container	Send to lab without delay	*1 hour
-Rectal swab	If indicated: proctoscopy, sigmoidoscopy	Insert swab 1 inch beyond anal sphincter and rotate once	One swab each on 3 consecutive days	Swab in Amies transport medium	Inferior to fresh stool. Adequate only for sigmoidoscopy specimens.	*2 hours
GENITAL TRACT FEMALE						
-Amniotic fluid		Aspirate with syringe	3-5 mL	Sterile container or syringe with needle replaced by a sterile Luer tip cap	Send to lab without delay	*1 hour
-Cervix	Wipe cervix clean of vaginal secretion and mucus. Use speculum without lubricants. Speculum may be rinsed with warm sterile water	Under direct vision gently compress cervix with blades of speculum and use a wringing motion with swab. Obtain exudate from endocervical glands	Endocervical secretion. Take 2 swabs if smears are to be made.	Swab in Amies transport medium See also R/O <i>Neisseria gonorrhoeae</i> in Table II.	Exceeding maximum swab transport time results in loss of gonococcal viability. Do not send dry or refrigerated swabs. See also R/O <i>Neisseria gonorrhoeae</i> in Table II.	*4 hours in Amies transport medium
-(For mycoplasma cultures)			Swab in Mycoplasma transport medium.		Obtain Mycoplasma transport medium in advance from Microbiology Lab. Place swab in medium immediately.	*4 hours in transport medium

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Specimen	Patient Preparation	Collection technique	Optimal specimen and volume	Collection container	Comments	Optimal Transport time
-Cul de sac	Surgical procedure	Aspiration through posterior vaginal vault	Fluid, secretion	Special anaerobic transport tube or syringe with needle replaced by sterile Luer tip cap	Send to lab without delay	*2 hours
-Chancre (Darkfield for <i>T. pallidum</i>)	1-2 hour soak with sterile saline and gauze	Scrape base of lesion, express fluid, avoid bleeding. Collect secretion into capillary tubes	Aspiration into capillary tubes	Capillary tubes	Test performed by STD clinic (telephone 731-3590)	* 30 minutes
-Endometrium	As for cervix	If swabs are to be used, collection through a sterile tube sheath may avoid contamination with vaginal flora	Curettings or aspiration	Special anaerobic transport tube or syringe with needle replaced by sterile Luer tip cap	Send to lab without delay	*1 hour
-Intrauterine device	Surgical	Surgical removal	Entire device plus secretion, pus	IUD in sterile anaerobe transport tube, or sterile urine cup if large IUD.	Send to lab without delay	*1 hour
-Products of conception (fetal, placenta, membranes)	Surgical	Select relevant or suspicious areas of tissues	Tissue or aspirates	Sterile container or syringe with needle replaced by sterile Luer tip cap	Send to lab without delay	*1 hour
-Urethra	Wipe clean with sterile gauze or swab	1 hour or more after urinating, "milk" the urethra or use thin wire swabs to collect material from 2 cm inside urethra	Swab with urethral secretion	Swab in Amies transport medium. See also Table II R/O <i>Neisseria gonorrhoeae</i>	Exceeding maximum swab transport time results in loss of gonococcal viability. Do not send dry or refrigerated swabs. See also Table II R/O <i>N. gonorrhoeae</i>	*4 hours in Amies

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Specimen	Patient Preparation	Collection technique	Optimal specimen and volume	Collection container	Comments	Optimal Transport time
-Uterus, tubes, ovaries	Usually surgical specimen	If surgical specimen, representative sample should be cut and submitted	Tissues, aspirates or swabs	Sterile container, syringe with needle replaced by sterile Luer tip cap, or special anaerobic swab transport tube.	Send to lab without delay	*1 hour
-Vagina		Simple aspiration or swabbing.	Aspirate or swab	Swab in Amies transport medium	See cervix. See also Table II R/O <i>N. gonorrhoeae</i> Cervix is preferred site for R/O <i>N. gonorrhoeae</i>	*4 hours in Amies
-Vulva (including labia, Bartholin's gland)	Skin prep as for regular skin sites. Do not use alcohol on mucous membranes	Collect with swab or aspirate with syringe and needle	Swab or aspirate (Bartholin's gland abscess)	Swab in Amies transport medium; aspirate in syringe with needle replaced by sterile Luer tip cap.		*4 hours in transport medium *2 hours in syringe
GENITAL TRACT						
MALE						
-Chancre (Darkfield)	See Chancre in female tract above					
-Penis	Skin prep as for regular skin sites. Do not use alcohol on mucous membranes.	Vesicles must be opened and secretion submitted as aspirate, or as a swab	Swab, aspirate, or secretions	Swab in Amies transport medium. Use Mycoplasma transport medium for genital Mycoplasma.	Do not send dry or refrigerated swabs.	*4 hours in Amies medium
-Prostate		Digital massage through rectum	Secretion	Sterile container or swab in Amies or Culturette transport medium. Use Mycoplasma transport medium for genital Mycoplasma	Helpful in some chronic urinary tract infections.	*4 hours in Amies transport medium; *2 hours in sterile cup

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-Urethra	Wipe clean with sterile gauze and saline	Same as for female; thin urethrogenital calcium alginate swabs are preferred	Swab with urethral secretion	Swab in Amies transport medium. Use Mycoplasma transport medium for genital Mycoplasma	In males the diagnosis of gonorrhea can often be made by microscopic examination of the gram-stained smear. See Penis above. See Table II, R/O <i>N. gonorrhoeae</i>	*4 hours in Amies transport medium
LYMPH NODES	Surgery	Aspiration, biopsy or excision	Tissue or aspirate	Sterile container or syringe with needle replaced by a sterile Luer tip cap	Indicate unusual organisms (e.g., LGV, viruses, <i>Actinomyces</i> , <i>Yersinia</i> , acid-fast bacteria) on request form.	*1 hour
ORAL CAVITY						
-Mucosal surface	Rinse mouth	Scrape with swab	Scrapings on swabs	Swab in Amies transport medium. For <i>M. pneumoniae</i> culture use Mycoplasma transport medium.	Indicate suspected organism. Smears are most useful for diagnosis of Vincent's angina or yeast infections.	*4 hours
-Dental abscess	Rinse mouth, prep with dry sterile gauze	Aspiration in syringe is preferable to swab	Fluid, pus	Syringe with needle replaced by a sterile Luer tip or anaerobic transport tube.	Send to lab without delay.	*2 hours
PUS (abscesses)	Skin decontamination	Aspiration in syringe preferable to swab.	As much as possible	Syringe with needle replaced by sterile Luer tip cap or anaerobic transport tube	Send to lab without delay. See dental abscess above.	*1 hour
RESPIRATORY TRACT						
-Bronchotracheal secretions or washings	Surgical or bronchoscopy	Intubation or transcutaneous aspiration brushings, tissue	3-5 mL Secretions, bronchial	Sterile container. Use Mycoplasma transport medium for <i>M. pneumoniae</i>	Indicate if quantitative culture is required. Place brush in 1.0 mL broth (obtain from Micro Lab) for quantitative brush culture.	*2 hours

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-Epiglottitis		Tongue blade and swab (See comments)	Swab	Culturette or swab in Amies transport medium	In cases of acute epiglottitis do not swab throat or use tongue blade unless prepared for tracheostomy.	*4 hours
-Lung	Surgical	Percutaneous or open lung surgery	Tissue or aspirated fluid	See Tissue below. Sterile tube or container.		*1 hour
-Nasopharynx		Pass thin wire swab gently through nostril into Nasopharynx. Stay near septum and floor of nose.	Swab	Thin wire swab in Culturette or Amies transport medium. Use Mycoplasma transport medium for <i>M. pneumoniae</i> .		*4 hours
-Nose		Rotate swab in anterior nares	Swab.	Swab in Amies transport medium	Used to R/O carriage of <i>S. aureus</i> . or MRSA. Do not send dry swab.	*4 hours
Sputum	Careful instruction of patient to produce lung material, not saliva	Expectoration. Assisted by saline nebulisation, postural drainage, as needed. Respiratory therapist may assist.	Morning specimen from deep cough, not saliva.	Sterile container. Use Mycoplasma transport medium for <i>M. pneumoniae</i> .	If microscopic exam reveals contamination with saliva, specimen will be rejected.	*2 hours
Throat	Tongue blade to depress tongue	Swab inflamed areas, collect pus or piece of membrane	Swab	Swab in Amies transport medium. Use Mycoplasma transport medium for <i>M. pneumoniae</i> .	See also R/O <i>Strep</i> , group A in Table II.	*4 hours
Tracheal aspirate		Through nose or mouth using sterile tube	Aspirate 1-2 mL	Sterile container. Use Mycoplasma transport medium for <i>M. pneumoniae</i> .		*2 hours

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Specimen	Patient Preparation	Collection technique	Optimal specimen and volume	Collection container	Comments	Optimal Transport time
R/O requests	See Table II page 9-25					
SKIN	Clean skin with 70% alcohol	Open vesicle, aspirate or swab or biopsy; punch biopsy for quantitative wounds	Needle aspirate, biopsy or swab	Syringe with needle replaced by sterile Luer tip cap, sterile container for biopsy, swab in Amies transport medium.	Do not add any fluids. Quantitative biopsies should be received at HMC Micro Lab before 1 PM Monday through Friday.	*1 hour-biopsy *2 hours-syringe *4 hours-swab
TISSUE	Surgery	Surgery or needle biopsy	1 mg piece desirable	Sterile container for large piece ($\geq 1\text{cm}^3$); anaerobic transport tube for small piece	Do not add any fluids. Send to lab without delay . Quantitative biopsies should be received at HMC Micro Lab before 1 PM Monday through Friday.	*1 hour
URINE -Clean voided urine	Careful instruction or assistance. Follow standard protocol in nursing care manual.	Clean voided mid-stream technique: void, discard, then void & collect.	First morning urine; several mL	Sterile wide-mouthed container with tight-fitting lid	May be refrigerated or collected in vacuum tubes containing preservative.	*24 hours in preservative tube. *4hours refrigerated. *1 hour at room temperature
-Catheter urine (indwelling catheter)		Iodine prep of aspiration site on catheter	Same as above	Sterile container	As above. Indicate catheterized specimen on requisition.	As above
-Suprapubic (bladder) urine	Surgical	Suprapubic aspiration	Same as above	Sterile container	As above. Indicate suprapubic collection on requisition.	As above

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Specimen	Patient Preparation	Collection technique	Optimal specimen and volume	Collection container	Comments	Optimal Transport time
WOUNDS	Surface decontamination	Swab active margin or obvious pus	Coat swab well	Culturette, Amies transport medium, or anaerobic transport tube	See ANAEROBIC CULTURE above for best recovery of anaerobic organisms.	*4 hours routine transport media; *2 hours anaerobic transport medium

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App. R - Microbiology Specimen Collection-Table II

TABLE II - R/O requests - **Indicate suspected organism on requisition so appropriate techniques are utilized**

Organism	Optimal specimen	Container	Comment	Maximum Transport time
<i>Bartonella (Rochalimaea)</i> species	Blood	10 mL blood in Isolator blood culture tube	Notify Micro lab in advance so Isolator tube can be obtained.	*4 hours
<i>Clostridium difficile</i>	Stool	Clean container with tight fitting lid	Toxin B test: send to SPS Antigen & Toxin A: send to Micro	*2 hours
<i>Corynebacterium diphtheriae</i>	Throat swab, wound swab	Swab in Amies transport medium		*4 hours
<i>E. coli</i> 0157:H7	Stool	Clean container with tight fitting lid		*2 hours
<i>Francisella tularensis</i>	lymph node drainage; blood	Drainage in sterile tube or syringe with luer tip cap. Blood in Isolator blood culture tube.	Notify Micro Lab in advance for optimal handling	*1 hour
<i>Haemophilus ducreyi</i>	Swab from base of ulcer inoculated on <i>H. ducreyi</i> medium at bedside	Direct inoculation of <i>H. ducreyi</i> medium is preferable to Culturette or Amies transport medium.	Notify Micro Lab in advance so media can be obtained	*1 hour
<i>Helicobacter (Campylobacter) pylori</i>	Biopsy from stomach or esophagus	Sterile tube	Notify Micro Lab in advance for optimal handling	*1 hour
<i>Legionella</i> species a. culture b. direct FA	Bronchial washing, lung biopsy or sputum	Sterile tube or Sterile container		*2 hours
MRSA (methicillin resistant <i>Staphylococcus aureus.</i>)	Anterior nares swab	Swab in Amies transport medium	Indicate R/O MRSA on requisition.	*2 hours
<i>Mycoplasma pneumoniae</i> culture	Throat swab or Lower Respiratory	Mycoplasma transport medium available in Micro Lab		*4 hours
<i>Mycoplasma</i> (Ureaplasma) culture only	Urogenital	Mycoplasma transport medium available in Micro Lab		*4 hours
<i>Mycoplasma</i> species culture only	Any specimen except blood	Mycoplasma transport medium available in Micro Lab		*4 hours

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Specimens beyond their maximum transport time should be recollected

App. R - Microbiology Specimen Collection-Table II

TABLE II - R/O requests - **Indicate suspected organism on requisition so appropriate techniques are utilized**

Organism	Optimal specimen	Container	Comment	Maximum Transport time
<i>Neisseria gonorrhoeae</i> ("GC") culture	Swab from urethra, cervix, vagina, throat, rectum.	Swab in Amies transport medium, or selective chocolate agar inoculated by clinician and incubated at 35°C with increased CO ₂	Dry or refrigerated swabs will not be cultured. Preinoculated agar should be maintained in increased CO ₂ preferably at 35°C, before transport to lab.	*Amies: 4 hrs Preinoculated agar not in CO ₂ : 30 min; 5 hrs if in CO ₂
	Joint fluid	Sterile container	Send to lab without delay	*2 hours
<i>Neisseria gonorrhoea</i> ("GC") nucleic acid detection by LCR (Note: Chlamydia LCR may be performed on same specimen.)	First void urine	Polypropylene (cloudy plastic) urine cup	Collect first voided 15 mL urine one or more hours after last void. Avoid blood, which may be inhibitory.	*2 days refrigerated; 4 days -20°C
	Cervical or urethral swab.	Abbott LCx swab collection kit available in lab.	Submit narrow LCx swab; discard large cleaning swab. Avoid blood, which may be inhibitory.	*2 days
Pertussis	Nasopharyngeal swab throat - ok.	Regan-Lowe transport tube available in Micro Lab	Notify Micro Lab in advance so transport tube can be obtained	*4 hours
<i>Staphylococcus aureus</i>	Anterior nares swab	Swab in Amies transport medium		*4 hours
<i>Strep</i> , group A beta-hemolytic culture	Throat swab, wound swab	Swab in Amies transport medium	For rapid results from throat specimens, specify group A Strep antigen test (Less sensitive than culture)	*4 hours
<i>Strep</i> , group A beta-hemolytic Rapid antigen detection	Throat swab	Swab in Culturette or <i>liquid</i> Amies transport medium. Do not send Amies gel transport medium.	Less sensitive than culture	*4 hours
<i>Strep</i> , group B beta-hemolytic	Vaginal and rectal swabs submitted together	Swab in Amies transport medium		*4 hours

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App. R - Microbiology Specimen Collection-Table III

TABLE III - Environmental Cultures

Organism	Optimal specimen	Container	Comment	Maximum Transport time
Autoclave sterility test	Spore strips or ampules	Submit sterile packet as is	Place in center of load. Clearly indicate location in autoclave	*8 hours
Fluids	1-3 mL or more	Sterile container	Follow FDA regulations for injectables	*4 hours
Surface area	Swab measured area, or press Rodac plate on surface.	Swab in Amies transport medium, or Rodac plate.		*4 hours

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Specimens beyond their maximum transport time should be recollected.

App. R - Microbiology Specimen Collection-Table IV

TABLE IV - Specimen Collection: Mycobacterial Culture and Acid-Fast Stains

Specimen	Specimen Requirements	Special Instructions
ASPIRATE	As much as possible in syringe with luer tip cap Dry swab is unacceptable.	Cleanse skin with alcohol before aspirating sample. Collect specimen on swab and place in transport media (Amies or Culturette) ONLY if volume is insufficient for aspiration by needle and syringe.
BIOPSY	1 gram of tissue, if possible, in sterile container	Collect aseptically and avoid indigenous flora. Select caseous portion if available. Do not immerse in saline or other fluid or wrap in gauze.
BLOOD	5 mL blood in Bactec Myco/F bottle	Disinfect site as for routine blood culture. Mix immediately.
BONE MARROW	As much as possible in YELLOW TOP SPS blood collection tube, or 1 to 5mL in Bactec Myco/F bottle.	Collect aseptically. Mix container immediately following collection.
BRONCHOSCOPY -Bronchoalveolar lavage (BAL)	5 mL, if possible, in sterile container.	Avoid contaminating bronchoscope with tap water; saprophytic AFB's may produce false positive culture and/or stain results.
-Bronchial brush	brush in sterile container	
BODY FLUIDS (pleural, pericardial, peritoneal, etc.)	Up to 400 mL, in sterile container or syringe with luer tip cap. Bloody specimens should be collected in YELLOW TOP SPS blood collection tube(s).	Smears are routinely done on body fluids and urines. Call Microbiology Lab to request exception. Fluids received with AFB Smear Only request will be cultured because smear without culture has unacceptably low sensitivity for mycobacteria detection.
CEREBROSPINAL FLUID	As much as possible in sterile container.	Smears are not routinely done for CSF. Call Microbiology Lab to request exception.
GASTRIC LAVAGE	5-10 mL in sterile container. Collect fasting early AM specimen on 3 consecutive days.	Use sterile saline to collect gastric lavage if sputum is not obtainable for AFB testing. Send specimen to lab <u>immediately</u> following collection; specimen delayed in transit >2 hours is unacceptable. Specimen will be neutralized on receipt in lab as AFB are destroyed by high acidity of gastric contents.
LYMPH NODE	Node or portion in sterile container without preservative or fixative	Collect aseptically and avoid indigenous flora. Select caseous portion if available. Do not immerse in saline or other fluid or wrap in gauze.

App. R - Microbiology Specimen Collection-Table IV

TABLE IV - Specimen Collection: Mycobacterial Culture and Acid-Fast Stains

Specimen	Specimen Requirements	Special Instructions
SKIN LESION	See Biopsy	Swabs in transport media (Amies or Culturette) are acceptable ONLY if biopsy or aspirate is not obtainable. Mycobacteria causing skin lesions have special growth requirements; therefore, we incubate duplicate cultures at 30°C, and 37°C. Hemin-containing medium is included for growth of <i>M. haemophilum</i> . <u>Cutaneous ulcer</u> : Collect biopsy from the periphery of the lesion or aspirate material from under the margin of the lesion. If skin lesion was acquired in tropical area, please note on request slip because <i>M. ulcerans</i> may require up to 3 months for primary isolation.
SMEARS ON SLIDES	Smear specimen over a 1.5 cm by 1.5 cm area of a clear slide.	Heat fix smears. Transport in a slide container that is taped closed and labeled BIOHAZARD.
SPUTUM	5-10 mL (1 mL minimum) in sterile container. Collect an <u>early AM</u> specimen from a deep productive cough on 3 consecutive days. <u>Do not pool</u> the above 3 specimens. Do not collect 24 hour sputum. Same day duplicate specimens are pooled. For follow-up of patients on therapy, collect at weekly intervals.	Avoid sputum contamination with nebulizer reservoir water. Saprophytic acid-fast bacilli in tap water may produce false positive culture and/or stain results. Induced sputum: Use sterile hypertonic saline. Indicate on request if specimen is an INDUCED sputum as these watery specimens resemble saliva and risk rejection as inadequate. If <1 mL sputum specimen, direct smear will be prepared and stained by the Auramine method and will not be processed for culture unless AFB are seen on smear. Negative culture results are unreliable for such small samples. Contact the Microbiology Lab to request an exception.
STOOL	5 to 10 grams in sterile container.	Direct smear for presence of AFB will be performed. If no AFB are seen, stool will not be cultured. If AFB are present, specimen will be decontaminated, concentrated, and cultured.
TISSUE	See Biopsy	
TRANSTRACHEAL ASPIRATE	As much as possible in syringe with luer tip cap or other sterile container.	
URINE	As much as possible (minimum 40 mL) first morning specimen, midstream or catheter in sterile container. Suprapubic tap: as much as possible, in syringe with luer tip or other sterile container.	Collect first morning specimen on three consecutive days. Only one specimen per day will be accepted. Do not collect 24 hour specimens. Smears will be performed on specimens collected by catheter or suprapubic tap only, as saprophytic acid-fast bacilli may colonize the urethra.
WOUND	See Biopsy	Swabs are not recommended

App. R - Microbiology Specimen Collection-Table V

TABLE V - Specimen Collection: Fungus Culture, KOH or India Ink Mounts

Specimen	Optimal Specimen	Container	Comments
ABCESS	Aspirate of abscess contents; as much as possible	Sterile container	Send to lab without delay
BIOPSY - See Tissue			
BLOOD	<u>Routine:</u> 5 mL blood	Collect directly Bactec MycoF/lytic bottle	Send to lab within 2 hours
	<u>R/O Histoplasma:</u> 10 mL blood.	Collect directly into Isolator tube (available in Micro Lab) after decontaminating tube top with iodine.	Send to lab without delay
BODY FLUIDS	As much as is available	Sterile container	Send to lab without delay
BONE MARROW	As much as is available	YELLOW-TOP (SPS) TUBE, or heparinized fluid,	Send to lab without delay
CENTRAL NERVOUS SYSTEM -Brain Biopsy	As much as is available	Sterile container	Send to lab without delay
-CSF	>2-3 mL	Sterile tubes	Send to lab without delay
EAR (otitis externa)	Scrapings or swab	Sterile tube	
EAR (otitis media or otitis interna)	Swab or aspirate	Sterile tube	For specimen from middle or inner ear use sterile funnel
EYE (corneal ulcer)	Use clean smooth spatula to obtain specimen	Directly inoculated fungal media or sterile tube	Contact laboratory if direct inoculation of media is desired.
GI TRACT (rectal, stool)	Swab	Amies transport medium or sterile container	Stool specimens are generally not cultured for fungus unless enterocolitis due to fungal overgrowth is suspected
ORAL CAVITY or TONGUE	Swab	Amies transport medium, or sterile tube	

App. R - Microbiology Specimen Collection-Table V

TABLE V - Specimen Collection: Fungus Culture, KOH or India Ink Mounts

Specimen	Optimal Specimen	Container	Comments
RESPIRATORY TRACT			
-Sputum	5-10 mL Early morning specimen is preferred.	Sterile container	Have the patient brush teeth and rinse mouth well with water just prior to collecting the specimen. Send to lab within 2 hours; if refrigerated, send within 6 hours.
-Tracheal aspirate	Expectorated or aspirated specimen from the lungs.	Sterile container	
-Lung Tissue	As much as possible	Sterile container	Send to lab without delay
-Nose and Nasopharynx	Swab	Sterile tube or Amies transport medium	
SKIN SYSTEM			
-Nail	Nail clippings, scrapings from active margin, and deep subungual debris. See comments.	Sterile container, petri dish	Disinfect the infected area with 70% alcohol and use sterile blade to scrape the nail or collect small pieces of the nail with sterile clipper. Superficial debris should be discarded before collection.
-Skin	Skin scrapings from active margin. See comments.	Sterile container or petri dish	After disinfecting the infected area with 70% alcohol and allowing the alcohol to dry, scrape from the periphery of one or more lesions. Include scrapings from the apparently healthy skin adjoining the lesion
-Hair	Pluck shaft and root of several infected hairs. If hair fluoresces under Wood's lamp, pluck the fluorescent hair.	Sterile container or petri dish	
-Vesicles	Snip off the tips of vesicles after disinfecting the vesicular area with 70% alcohol	Sterile container or petri dish	
TISSUE			
-Lymph nodes, -Biopsy	Biopsy material of walls and central area of tissue (as much as possible)	Sterile container with small amount of nonbacteriostatic saline or sterile broth to keep specimen moist.	Send to lab without delay.

App. R - Microbiology Specimen Collection-Table V

TABLE V - Specimen Collection: Fungus Culture, KOH or India Ink Mounts

Specimen	Optimal Specimen	Container	Comments
ULCERS	Saline aspirates from the ulcer base and wall. If biopsied, include aspirates of the ulcer and wall	Sterile container	A syringe with 23 or 25 gauge needle is convenient to use for collecting base aspirates. Send to lab without delay.
UROGENITAL SYSTEM			
-Urine	10-50 mL urine. First morning specimen is preferred	Sterile container	Send to lab within 1 hour; if refrigerated, deliver within 4 hours
-Prostatic secretions, -Cervix, vagina	Swab well-coated with secretions	Swab or secretions in sterile tube, or Amies transport medium	

App. R - Microbiology Specimen Collection-Table VI

Table VI - Specimen Collection: Parasites

Specimen	Parasite	Optimal Specimen	Collection	Comments
BLOOD	<i>Plasmodium</i> spp.	Full tube of anticoagulated blood (LAVENDER TOP [EDTA] or GREEN TOP [HEPARIN]).	Anytime	Collect blood before initiating treatment. Send blood to the laboratory as soon as possible. Thin smears will be examined and reported ASAP. One negative set of smears does not rule out malaria.
	<i>Trypanosoma</i> spp.	Full tube of anticoagulated blood (LAVENDER TOP [EDTA] or GREEN TOP [HEPARIN]).	During febrile episodes.	
	Microfilariae	Full tube of anticoagulated blood (LAVENDER TOP [EDTA] or GREEN TOP [HEPARIN]).	(<i>Wuchereria/Brugia</i>) Collect between 10 PM and 2 AM. (Others except for <i>Onchocerca</i>) Collect between 10 AM - 6 PM	
BONE MARROW	<i>Leishmania</i>	Bone marrow aspirate in anticoagulant.	Anytime	Cultures are available but only after prior arrangement with the Microbiology lab.
CSF (cerebrospinal fluid)	<i>Naegleria</i> sp. <i>Acanthamoeba</i> sp.	Freshly collected.		Do not refrigerate. Send to the laboratory immediately. Cultures are available only after prior arrangements with the Microbiology lab.
CYST	<i>Echinococcus</i>	Unprocessed cyst fluid.		Keep the biopsy moist in a minimum of sterile isotonic saline.
	<i>Entamoeba histolytica</i>	Cyst wall biopsy		Send to the laboratory immediately. Prior notification of the Microbiology lab will ensure priority service. Serologic testing may be useful.
EYE	<i>Acanthamoeba</i> sp.	Corneal scrapings in Pages saline	Suspend the corneal scrapings in approximately 1/2 cc sterile Pages saline.	Send to the laboratory immediately. Make prior arrangements with the Microbiology lab to have the specimen cultured for amoeba.

App. R - Microbiology Specimen Collection-Table VI

Table VI - Specimen Collection: Parasites

Specimen	Parasite	Optimal Specimen	Collection	Comments
FECES	All intestinal parasites (except <i>Cryptosporidium</i> , microsporidia or <i>Cyclospora</i> , see comments and below) nematodes and trematodes	A series of 3 stools collected in the AM either every other day or 3 consecutive days.	Clean container, two-vial preservative system (PVA & Formalin), or one vial system (Ecofix or Unifix) Collect all fecal specimens prior to administration of any medications, enemas, or cathartics	Send fresh specimens to the lab immediately. Do not refrigerate. Requests for R/O cryptosporidia, microsporidia or <i>Cyclospora</i> must be specified on the request form. See below for <i>R/O Giardia only</i>
	<i>Giardia</i> only	Single stool	<ul style="list-style-type: none"> Unpreserved or preserved in 10% formalin. (PVA, Ecofix and Unifix are unacceptable.) Refrigerate specimen 	<ul style="list-style-type: none"> Indicate <i>Giardia only</i> or <i>Giardia antigen</i> on requisition.
	<i>Cryptosporidium</i> sp. Microsporidia <i>Cyclospora</i> sp.	Single stool	Do not place the specimen in preservative solution. Refrigerate specimen	
SPUTUM	<i>Paragonimus</i> <i>Strongyloide</i>	Expectorated or induced sputum.	Fresh AM Specimen	Send to lab immediately. Up to 3 specimens each day is acceptable. Stool exams recommended if sputum is negative but symptoms persist.
MISCELLANEOUS Biopsies	<i>E. histolytica</i>	Freshly collected scrapings (cuvette, Volkman spoon or other surgical instrument.) About 1 mL aspirate/wash of mucosal lesion.	2 to 3 hours after a bowel movement or purge.	Send to the laboratory immediately. Prior notification of the Microbiology lab will ensure priority handling of the specimen. Do not use swabs to collect the specimen.
Bladder wall	<i>Schistosoma haematobium</i>	Biopsy from area around the Trigone.	Collect in clean container.	Send to the laboratory immediately.
Muscle	<i>T.aenia solium</i> <i>Trichinella spiralis</i>	Muscle biopsy.	Collect in clean container.	Send to the laboratory immediately.
Rectum	<i>Schistosoma. mansoni</i> <i>Schistosoma. japonicum</i>	Biopsy from the level of the dorsal fold (Houston valve) about 9 cm from the anus.	Collect in clean container.	Send to the laboratory immediately.

App. R - Microbiology Specimen Collection-Table VI

Table VI - Specimen Collection: Parasites

Specimen	Parasite	Optimal Specimen	Collection	Comments
MISCELLANEOUS Biopsies (continued) Skin	<i>Onchocerca volvulus</i>	African: "Skin snips" from thigh, buttocks or iliac crest American: "Skin snips" from scalp, face, buttocks.	Collect in clean container.	Send to the laboratory immediately.
	<i>Leishmania spp.</i>	Aseptically collect biopsy of advancing ulcer edge or ulcer bed.	Sterile jar with moist gauze.	Prior arrangement must be made with the Microbiology lab so that culturing material will be available. Send to the laboratory immediately.
"String Test" Enteric test	<i>Giardia</i> <i>Strongyloides</i>	"String"	Follow package insert instructions. Collect in clean container	Send to the Microbiology lab immediately. If <i>Giardia</i> is suspected, a Giardia antigen test is recommended.
Perianal "Scotch tape"/swab	<i>Enterobius vermicularis</i>	"Scotch tape" (clear cellophane tape).	Collect in AM before bathing or bowel movement. Alternatively, collect after patient has been resting/sleeping quietly for several hours.	If not familiar with this collection method, contact the laboratory for assistance. "Invisible" or frosted type is not acceptable. "Scotch-tape" is available in the micro lab.
Urogenital Vagina Urethra Prostatic secretion	<i>Trichomonas. vaginalis</i>	Sterile swabs or secretions in a tube with a small amount of sterile saline.	Sterile tube	Send to the laboratory immediately. Do not refrigerate. Do not douche within 3-4 days before collecting the specimen
Urine	<i>Schistosoma. haematobium</i>	Collect between Noon and 3 p.m. or collect a 24 hour specimen.	Use a clean container No preservatives.	If electing to collect a 24 hour sample, refrigerate the sample between collection periods. The last portion of each void specimen is the most important aliquot.
Insects/ Arthropods			<ul style="list-style-type: none"> • Use a clean container. • No Preservatives. 	Contact the Microbiology lab for instructions.
Intact worms or proglottids		"Passed/voided" specimen.	Clean jar with isotonic saline or water.	Do not use ethanol or formalin to preserve the specimen.