Bacteriology, Mycology, and Mycobacteriology Specimen Collection Guidelines

Specimen Type	Collection		Time and Temp		Replica Limits	Comment(s)
	Guidelines	Device and/or minimum volume	Local Transport[b]	Courier or local storage		
Abscess	Remove surface exudate by wiping with sterile saline or 70% ETOH[a].					Tissue or fluid is always superior to a swab specimen. If swabs must be used, collect 2: one for culture and one for Gram staining. Preserve them with Stuart's or Amies medium.
Open	Aspirate if possible, or pass a swab deep into the lesion and firmly sample the lesion's advancing edge.	ESwab transport system	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day/source	A sample from the base of the lesion and a sample from the abscess wall are most productive.
Closed	Aspirate abscess wall material with needle and syringe. Aseptically transfer all material into anaerobic transport device or sterile tube.	Transport Tube, ≥ 1 ml or sterile tube.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day/source	Sampling of the surface area can introduce colonizing bacteria not involved in the infectious process.
Bone marrow	Prepare puncture site as for surgical incision	Inoculate a pediatric l.5 ml. lysis centrifugation tube.	≤ 16 h, RT[a] if in culture bottle or tube.	≤ 16 h, RT[a]	1/day	Small volumes of bone marrow may be inoculated directly onto culture media.
Burn	Clean and debride the wound prior to specimen collection.	Tissue placed in a screw-cap container. Aerobic culture. Swab exudate.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day/source	A 3 to 4 mm punch biopsy is optimum when quantitative cultures are ordered. Process for aerobic culture only. Quantitative culture may or may not be valuable. Surface cultures of burns may be misleading.
Catheter: I.V.	1. Cleanse the skin around the catheter site with alcohol. 2. Aseptically remove catheter and clip a 5-cm distal tip of the catheter directly into a sterile tube. 3. Transport directly to microbiology to prevent drying.	Sterile screw-cap tube or cup	≤ 15 min., RT[a]	≤ 24 h, 4°C[a]	None	Acceptable i.v. catheters for semiquantitative culture (Maki method): central, CVP, Hickman, Broviac, peripheral, arterial, umbilical, hyperalimentation, Swan-Ganz.
Foley	Do not culture since growth represents distal urethral flora.					Not acceptable for culture.
Cellulitis	1. Cleanse site by wiping with sterile saline or 70% alcohol. 2. Aspirate the area of maximum inflammation (commonly the center rather than the leading edge) with a fine needle and syringe. 3. Draw small amount of sterile saline into syringe and aspirate into sterile screw-cap tube.	Sterile tube (syringe transport not recommended)	≤ 15 min., RT[a]	≤ 24 h, RT[a]	None	Yield of potential pathogens is only 25-35%.

[a] ETOH = ethanol; RT = room temperature; i.v. = intravenous; AFB = acid-fast bacilli; CSF = cerebrospinal fluid; BAP = blood agar plate; CHOC = chocolate agar; BHI = brain heart infusion; PVA = polyvinyl alcohol fixative, 4ºC = refrigeration.

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CSF	1. Disinfect site with 2% iodine tincture. 2. Insert a needle with stylet at L3-L4, L4-L5, or L5-S1 interspace. 3. On reaching the subarachnoid space, remove the stylet and collect 1-2 ml. of fluid in each of three leakproof tubes.	Sterile screw-cap tube Minimum amount required: bacteria, ≥ 1 ml; fungi, ≥ 2 ml; AFB[a], ≥ 2 ml; virus, ≥ 1 ml;	Bacteria: never refrigerate; ≤ 15 min, RT[a] Virus: transport on ice; ≤15 min, 4°C[a]	≤ 24 h, RT[a] ≤ 72 h, 4°C[a]	None	Obtain blood cultures also. If only 1 tube of CSF[a] is collected, it should be submitted to microbiology first; otherwise submit tube 2. Aspirate of brain abscess or a biopsy may be necessary to detect anaerobic bacteria or parasites.
Decubitus ulcer	See comment: A swab specimen is not the specimen of choice. 1. Cleanse surface with sterile saline. 2. If a sample biopsy is not available, vigorously swab the base of the lesion. 3. Place the swab in appropriate transport system.	ESwab or Transport Tube, or sterile tube (for tissue)	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day/source	A decubitus swab provides little clinical information; discourage collection of it. A tissue biopsy sample or a needle aspirate is the specimen of choice.
Ear: Inner	Tympanocentesis should be reserved for complicated, recurrent, or chronic persistent otitis media. 1. For an intact ear drum, clean the ear canal with soap solution and collect fluid via the syringe aspiration technique 2. For a ruptured ear drum, collect fluid on a flexible-shaft swab via an auditory speculum.	Sterile tube, or Transport Tube.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day/source	Throat or nasopharyngeal cultures are not predictive of agents responsible for otitis media and should not be submitted for that purpose.
Outer	1. Use a moistened swab to remove any debris or crust from the ear canal. 2. Obtain a sample by firmly rotating the swab in the outer canal.	ESwab transport system.	≤ 2 h, RT[a]	≤ 24 h, 4°C[a]	1/day/source	For otitis externa, vigorous swabbing is required since surface swabbing may miss streptococcal cellulitis.
Eye: Conjunctiva	1. Sample both eyes using separate swabs (pre-moistened with sterile saline) by rolling over each conjunctiva. 2. Inoculate media at time of collection. 3. Smear swabs onto 2 slides for staining.	Direct culture inoculation: BAP[a] and CHOC[a] or ESwab transport system.	Plates: ≤ 15 min. RT[a] Swabs: ≤ 2 h, RT[a]	≤ 24 h, RT[a]	None	If possible, sample both conjunctivae, even if only one is infected, to determine the indigenous microflora. The uninfected eye can serve as a control with which to compare the agents isolated from the infected eye.
Corneal Scrapings	1. Obtain conjunctival swab specimens as described above. 2. Instill 2 drops of local anesthetic. 3. Using a sterile spatula, scrape ulcers or lesions and inoculate scraping directly onto media. 4. Apply remaining material to 2 clean glass slides for staining.	Direct culture inoculation, blood, CHOC[a], and SAB[a] agar	≤ 15 min. RT[a]	≤ 24 h, RT[a]	None	It is recommended that swabs for culture be taken prior to anesthetic application, whereas corneal scrapings can be obtained afterward.

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Fluid or aspirates	Prepare eye for needle aspiration of fluid.	Sterile screw-cap tube or direct inoculate of small amount of fluid onto media.	≤15 min. RT[a]	≤ 24 h, RT[a]	1/day	Include fungal media. Anesthetics may be inhibitory to some etiologic agents.
Feces: Routine culture	1. Pass directly into a clean, dry container. 2. Transport the specimen to lab within 1 h of collection or transfer a portion to a Cary-Blair transport system. 3. Cary-Blair: carefully remove the cap and attached spoon to pick several spoonfuls of the stool, especially from areas that are slimy, bloody, or watery. Place the stool into the vial to the fill line. Mix well.	Clean, leak-proof, wide-mouth container or a Cary-Blair transport system; ≥ 2 g	Unpreserved: ≤ 1 h, RT[a] Cary-Blair transport system: ≤ 24 h, RT[a]	≤ 24 h, 4°C[a] ≤ 48 h, RT[a] or 4°C[a]	1/day	Do not perform routine stool cultures on patients whose length of stay was > 3 days and the admitting diagnosis was not gastroenteritis. Culture and toxin tests for Clostridium difficile should be considered in these cases. Swabs for routine pathogens are not recommended except in infants and in patients with active diarrhea (see Rectal swabs).
C. difficile	Pass liquid or soft stool directly into a clean, dry container. Soft stool is defined as stool assuming the shape of its container. A swab specimen is not recommended for toxin testing.	Sterile, leak-proof, wide-mouth container; ≥ 5 ml	≤1 h, RT[a]; 1-24 h, 4°C[a] > 24 h, -20°C	2 days, 4°C[a], for culture 3 days, 4°C[c], or longer at –70°C for toxin test	1 /2 days	Patients should be passing ≥ 5 liquid or soft stools per 24 h. Testing of formed or hard stool is often unproductive and may indicate only commensal carriage.
Escherichia coli For Shiga-like (Vero) toxin (0157 and other serotypes)	Pass liquid or bloody stool into a clean, dry container.	Sterile, leak-proof wide-mouth container or Cary-Blair transport system	Unpreserved: ≤1 h, RT; Cary-Blair transport system < 24h, RT[a] or 4°C[a]	≤ 24 h, 4°C[a] ≤ 48 h, RT[a]	1/day	Bloody or liquid stools collected within 6 days of onset from patients with abdominal cramps have the highest yield.
Leukocytes	Pass feces directly into a clean, dry container. Transport specimen to lab within 1 h of collection or transfer to ova and parasite transport system (SAF).	Sterile, leak-proof, wide-mouth container > 2 ml	Unpreserved: ≤ 1 h, RT[a]	≤ 24 h, 4°C[a] Indefinite, RT[a]	1/day	
Rectal swab	1. Carefully insert a swab ≈ 1 in. beyond the anal sphincter. 2. Gently rotate the swab to sample the anal crypts. 3. Feces should be visible on the swab for detection of diarrheal pathogens.	ESwab transport system.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	Reserved for detecting Neisseria gonorrhoeae, Shigella spp., Campylobacter spp., HSV, and anal carriage of group B Streptococcus spp. or for patients unable to pass a specimen.
Fistulas	See Abscess					

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Fluids: abdominal, amniotic, ascites, bile, joint, paracentesis, pericardial, peritoneal, pleural, synovial, thoracentesis	1. Disinfect overlying skin with 2% iodine tincture. 2. Obtain specimen via percutaneous needle aspiration or surgery. 3. Transport specimen to laboratory immediately. 4. Always submit as much fluid as possible; *never* submit a swab dipped in fluid.	Blood culture bottle for bacteria and yeast or sterile screw-cap tube or Transport Tube. Bacteria, ≥ 1 ml; fungi, ≥ 10 ml; mycobacteria, ≥ 10 ml	≤15 min, RT[a]	≤ 24 h, RT[a] Pericardial fluid and fluids for fungal cultures: ≤ 24 h, 4°C[a]	None	Amniotic and culdocentesis fluids should be transported in anaerobic system and need not be centrifuged prior to Gram staining. Other fluids are best examined by Gram staining of a cytocentrifuged preparation. See Table 4.
Gangrenous tissue	See Abscess					Discourage sampling of surface or superficial tissue; tissue biopsy or aspirates are preferred.
Gastric: wash or lavage	Collect early in the morning before patients eat and while they are still in bed. 1. Introduce a nasogastric tube orally or nasally into the stomach. 2. Perform lavage with 25-50 ml of chilled, sterile, distilled water. 3. Recover sample and place it in a leakproof, sterile container. 4. Before removing the tube, release suction and clamp it.	Sterile, leak-proof container.	≤ 15 min, RT[a]	≤ 24 h, 4°C[a]	1/day	The specimen must be processed promptly since mycobacteria die rapidly in gastric washings.
Genital (female): Amniotic	1. Aspirate via amniocentesis, cesarean section, or intrauterine catheter.	Transport Tube or sterile tube, ≥ 1 ml	≤ 15 min, RT[a]	≤ 24 h, RT[a]	None	Swabbing or aspiration of vaginal membrane is *not* acceptable because of the potential for culture contamination by commensal vaginal flora.
Bartholin	1. Disinfect skin with a iodine preparation. 2. Aspirate fluid from ducts.	Transport Tube ≥ 1 ml	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	
Cervical	1. Visualize the cervix using a speculum without lubricant. 2. Remove mucus and secretions from the cervix with a swab and discard the swab. 3. Firmly, yet gently, sample the endocervical canal with a newly obtained sterile swab.	ESwab transport system. GC Selective plate.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	See information on virus and chlamydia collection and transport needs. Neisseria gonorrhoeae is found in exudates, whereas chlamydiae infect specific cells.
Cul-de-sac	Submit aspirate or fluid	Transport Tube > 1 ml	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	

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Endometrial	1. Collect transcervical aspirate via a telescoping catheter. 2. Transfer the entire amount to an anaerobic transport system.	Transport Tube ≥ 1 ml	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	
Products of conception	1. Submit a portion of tissue in a sterile container. 2. If obtained by cesarean section, immediately transfer it to an anaerobic transport system	Sterile tube or Transport Tube	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	Do not process lochia. Culture of this specimen may or may not provide clinically relevant results, and such results can be misleading.
Urethral	Collect 1h after patient has urinated. 1. Remove exudate from the urethral orifice. 2. Collect discharge material on a swab by massaging the urethra against the pubic symphysis through the vagina.	ESwab transport system.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	If no discharge can be obtained, wash the external urethra with Betadine soap with rinse with water. Insert a urethrogenital swab 2-4 cm. into the urethra; rotate swab for 2 s.
Vaginal	1. Wipe away an excessive amount of secretion or discharge. 2. Obtain secretions from the mucosal membrane of the vaginal vault with a sterile swab or pipette. 3. If a smear is also requested, use a second swab.	ESwab transport system.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	For intrauterine devices, place entire device in a sterile container and submit at RT. Gram staining is recommended for confirmation of bacterial vaginosis. Results from cultures are often inaccurate and misleading.
Genital (female or male): Lesion	1. Clean the lesion with sterile saline and remove the surface of the lesion with a sterile scalpel blade. 2. Allow transudate to accumulate. 3. Pressing the base of the lesion, *firmly* sample exudate with a sterile swab.	ESwab transport system.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	Specimens for syphilis should not be submitted for culture.
Genital (male): Prostate	1. Cleanse the glans with soap and water. 2. Massage the prostate through the rectum. 3. Collect fluid on a sterile swab or in a sterile tube.	ESwab transport system or sterile tube	≤ 2 h, RT[a]	≤24 h, RT[a]	1/day	More relevant results may be obtained by adding a urine specimen immediately before and after massage to indicate urethral and bladder organisms. Ejaculate can also be cultured.
Urethra	Insert a urethrogenital swab 2-4 cm. into the urethral lumen, rotate the swab, and leave it in place for at least 2 s to facilitate absorption.	ESwab transport system. GC selective place	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	See information on virus and chlamydia collection and transport needs.

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Hair: dermatophytosis	1. With forceps, collect at least 10-12 affected hairs with the base of the shaft intact. 2. Place in a clean tube or container.	Clean container, 10 hairs	≤ 24 h, RT[a]	1/day/site		Collect scalp scales, if present, along with scrapings of active borders of lesions. Note any antifungal therapy taken recently.
Nail: dermatophytosis	1. Wipe the nail with 70% alcohol using gauze (not cotton). 2. Clip away a generous portion of the affected area and collect material or debris from under the nail. 3. Place material in a container.	Clean container, enough scrapings to cover the head of a thumb tack.	≤ 24 h, RT[a]		1 day	
Pilonidal cyst	See Abscess					
Respiratory, lower: Bronchoalveolar lavage, bronchial brush or wash, tracheal aspirate	1. Place aspirate or washing in a sputum trap. 2. Place brush in a sterile container with saline.	Sterile container, > 1 ml	≤ 2 h, RT[a]	≤ 24 h, 4°C[a]	1/day	
Sputum, expectorate	1. Collect the specimen under the direct supervision of a nurse or physician. 2. Have the patient rinse or gargle with water to remove superficial flora. 3. Instruct the patient to cough deeply to produce a lower respiratory specimen (not postnasal fluid). Collect in a sterile container.	Sterile container, > 1 ml Minimum amounts: Bacteria, > 1 ml; fungi, 3-5 ml; mycobacteria, 5-10 ml; parasites, 3-5 ml	≤ 2 h, RT[a]	≤ 24 h, 4°C[a]	1/day	For pediatric patients unable to produce a specimen, a respiratory therapist should collect a specimen via suction. The best specimen should have ≤ 10 squamous cells per 100 x field.
Sputum, induced	1. Have the patient rinse mouth with water after brushing the gums and tongue. 2. With the aid of a nebulizer, have the patient inhale ≈ 25 ml of 3-10% sterile saline. 3. Collect the induced sputum in a sterile container.	Sterile container	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	*Histoplasma capsulatum* and *Blastomyces dermatitidis* survive for only short periods of time once a specimen is obtained. Fungal recovery is primarily for *Cryptococcus* spp. and some filamentous fungi; other yeasts rarely cause lower respiratory tract infection.

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Respiratory, upper: Oral	1. Remove oral secretions and debris from the surface of the lesion with swab and discard swab. 2. Using a second swab, vigorously sample the lesion, avoiding any areas of normal tissue.	ESwab transport system.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	Discourage sampling of superficial tissue for bacterial evaluation. Tissue biopsy or needle aspirates are the specimens of choice.
Nasal	1. Insert a swab, pre-moistened with sterile saline ≈ 2 cm into the nares. 2. Rotate the swab against the nasal mucosa.	ESwab transport system.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	Anterior nose cultures should be reserved for detecting staphylococcal and streptococcal carriers or for nasal lesions.
Throat	1. Depress the tongue with a tongue depressor. 2. Sample the posterior pharynx, tonsils, and inflamed areas with a sterile swab.	ESwab transport system.	≤ 2 h, RT[a]	≤ 24 h, RT[a]	1/day	Throat cultures are contraindicated for patients with an inflamed epiglottis. Swabs for N. gonorrhoeae should be inoculated or GC selective plates and transport at RT.
Skin: Dermato-phytosis	1. Cleanse the affected area with 70% alcohol. 2. Gently scrape the surface of the skin at the active margin of the lesion. *Do not draw blood.* 3. Sample the posterior pharynx, tonsils, and inflamed areas with a sterile swab.	ESwab transport system.	≤ 24 hr, RT[a]		1/day/site	If the specimen is submitted between glass slides, tape the slides together and submit in an envelope.
Tissue	1. Submit in a sterile container. 2. For small samples, add several drops of sterile saline to keep moist. 3. *Do not allow tissue to dry out*. 4. Place in a sterile, moist jar.	Sterile, screw-cap jar. Saline may need to be added.	≤ 15 min, RT[a]	≤ 24 h, RT[a]	None	Always submit as much tissue as possible. If possible, save an amount of surgical tissue at –70°C in case further studies are needed.
Urine: Female, midstream	1. Thoroughly cleanse the urethral area with soap and water. 2. Rinse the area with wet gauze pads. 3. While holding the labia apart, begin voiding. 4. After several milliliters have passed, collect a midstream portion without stopping the flow of urine.	Sterile wide-mouth container, ≥ 1 ml, or urine transport kit	Unpreserved: ≤ 2 h, RT[a] Preserved: ≤ 24 h, RT[a]	≤ 24 h, 4°C[a]	1/day	

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Urine Male, midstream	1. Cleanse the glans with soap and water. 2. Rinse with wet gauze pads. 3. Holding the foreskin retracted, begin voiding. 4. After several milliliters have passed, collect a midstream portion without stopping the flow of urine.	Sterile wide-mouth container, ≥ 1 ml, or urine transport kit	Unpreserved: ≤ 2 h, RT[a] Preserved: ≤ 24 h, RT[a]	≤ 24 h, 4°C[a]	1/day	
Straight catheter	1. Thoroughly cleanse the urethral area with soap and water. 2. Rinse the area with wet gauze pads. 3. Aseptically, insert a catheter into the bladder. 4. After allowing ≈ 15 ml to pass, collect urine to be submitted in a sterile container.	Sterile, leakproof container	Unpreserved: ≤ 2 h, RT[a] Preserved: ≤ 24 h, RT[a]	≤ 24 h, 4°C[a]	1/day	If preparation is inadequate, the procedure may introduce urethral flora into the bladder and increase the risk of iatrogenic infection.
Indwelling catheter	1. Disinfect the catheter collection port with 70% alcohol. 2. Use a needle and syringe to aseptically collect 5-10 ml of urine. 3. Transfer to a sterile tube or container or urine transport system.	Sterile, leakproof container or urine transport kit	Unpreserved: ≤ 2 h, RT[a] Preserved: ≤ 24 h, RT[a]	≤ 24 h, 4°C[a]	1/day	
Wound	See Abscess					

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Revised July 2015