

Blood Cultures - Aerobic & Anaerobic

Blood cultures are drawn into special bottles that contain a special medium that will support the growth and allow the detection of micro-organisms that prefer oxygen (aerobes) or that thrive in a reduced-oxygen environment (anaerobes).

Multiple samples are usually collected. Routine standard of care indicates a minimum of Two Separate Sites or collected At Least 15 Minutes Apart. Multiple Collection/Multiple Site collection is done to aid in the detection of micro-organisms present in small numbers and/or may be released into the bloodstream intermittently.

Samples must be incubated for several days before resulting to ensure that the sample is indeed negative before resulting out. Synergy Laboratories uses state of the art automated instrumentation that provides continuous monitoring. This allows for more rapid detection of samples that do contain bacteria or yeast.

CONSIDER AEROBIC BLOOD CULTURES IN:	CONSIDER ANAEROBIC BLOOD CULTURES IN:
1) New onset of fever, change in pattern of fever or unexplained clinical instability.	1) Intra-abdominal infection
2) Hemodynamic instability with or without fever if infection is a possibility.	2) Sepsis/septic shock from GI site
3) Possible endocarditis or graft infection.	3) Necrotizing fasciitis or complicated skin/soft tissue infection.
4) Unexplained hyperglycemia or hypotension.	4) Severe oropharyngeal or dental infection.
5) To assess cure of bacteremia.	5) Lung abscess or cavitary lesion.
6) Presence of a vascular catheter and clinical instability.	6) Massive blunt abdominal trauma.

Body Fluid Specimens

Selection

- Collect the specimen using strict aseptic technique.

- The patient should be fasting.
- If only one tube of fluid is available, the microbiology laboratory gets it first. IF more than one tube (1ml each) is available, microbiology should get the second or third tube, whichever is less bloody.
- Draw CSF at L3-L4 or lower to avoid spinal cord damage. Draw it at L4-L5 in children, because the conus medullaris extends lower in children than in adults.

Collection Materials

- CSF tray
- Skin disinfectant
- Sterile towels or drape
- Novocaine (0.5 to 1 %), needle, syringe
- Two lumbar puncture needles, small bore (20 to 22 gauge) with stylet . Water manometer
- Three small, sterile, screw-cap tubes

Collection Method

- Ensure that the patient will be motionless during the procedure. Restraints may be necessary.
- Explain that some pain is inevitable: local anesthesia rarely reaches the meninges, and pain occurs when the needle stretches the dura and pulls on connective tissue around the vertebrae.
- Have the patient arch his or her back so that the head almost touches the knees.
- Disinfect the skin along a line drawn between the crests of the two ilia if a puncture is to be made in the lumbar region.
- The physician introduces the needle. A stylet is used to avoid implantation of skin, which may cause dermoid cysts to form in the spinal canal.
- As soon as the fluid starts to drop from the needle, the needle's position in the subarachnoid space is established.
- Measure the pressure of the CSF at this point.
- Collect the drops of fluid (as much as 1 ml if possible) into sterile, screw-cap tubes.

Labeling

- Label the specimen with patient information.
- Indicate the age of the patient on the requisition.
- Indicate any therapy being given.

Transport

- Do not refrigerate the specimen.
- Hand carry the specimen to the laboratory.

Comments

- A Gram stain of the fluid should be done and the results read as soon as possible.
- It is essential that any culture and stain information about CSF be immediately telephoned or given to the physician so that therapy can be evaluated early. The age of the patient is a clue to the technologist as to the possible agent causing the illness

HERPES SIMPLEX VIRUS - PCR

Collection of CSF

- Aseptically collect CSF into appropriate sterile tubes CSF tubes
- Properly label tubes as collecting, or pre-label. Tubes labeled improperly may impact quality and interpretation of test results. See table below:

Tube 1	Cell count
Tube 2	Stat gram stain and culture (C+S) (2 cc.)
Tube 3	Glucose and protein
Tube 4	Cell count (for comparison to Tube 1)
Tube 5 (optional)	Virology, mycology, cytology, etc.

- Inpatient: Transport a minimum of 1 mL of CSF to laboratory on ice at 2-8°C.

Bronchoalveolar lavage (BAL), ocular fluid, amniotic fluid

- Aseptically collect and transfer 1 mL (minimum) of fluid to a sterile container/transport tube.
- A heparinized (green/no gel) or plain no-additive (red/no gel) Vacutainer® or Greiner® evacuated collection tube can be used for transport.
- Inpatient: Transport to laboratory on ice at 2-8°C.
- Outreach: Transport to laboratory refrigerated

Plasma or Serum

- Collect One 7 mL plain red or serum separator tube (serum) or Two 5 mL lavender (EDTA) or One 7mL Pink (EDTA)
- Inpatient: Send to Lab ASAP for processing
- Outpatient/Outreach: Spin blood specimen and remove serum or plasma before transporting. *Specimen source is required. *Note "PLASMA" or "SERUM" on tube and order requisition. Transport to laboratory refrigerated
- Vesicle fluid

- Vesicle fluid should be collected on sterile swab and transported in viral transport media (Microtest M4 or UTM)
- Outpatient/Outreach: Transport to laboratory refrigerated

Biopsy tissue

- Snap frozen and send on dry ice or in viral transport media (Microtest M4 or UTM)
- Inpatient: Send to Lab for processing
- Outpatient/Outreach: Transport to laboratory refrigerated

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HSV by PCR COLLECTION SUPPLIES

Lumber Puncture Numbered Tubes



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Heparinized (green/no gel)

Plain no-additive (red/no gel)



Serum separator tube (Gold)



EDTA (Lavender)



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Microtest M4 (UTM)



Cervical or Endocervical Specimens

Selection

- Contamination of cervical or endocervical specimens with vaginal secretions will interfere with the recovery of *N. gonorrhoea* and invalidate interpretation by Gram stain.
- Select only material from the endocervix by using a speculum to aid in seeing the area

Collection Materials

- Cervical speculum
- Swabs
- Transport medium
- Warm water

Collection Method

- Moisten the speculum with warm water. Lubricants can be toxic to *Neisseria*.
- Gently compress the cervix with the blades of the speculum, and collect the endocervical discharge with a calcium alginate, Dacron, or non-toxic cotton swab. Alternatively, insert the swab into the cervical and allow the swab to remain in place for a few seconds, and remove it.
- Anal cultures may be collected to accompany cervical specimen when *N. gonorrhoea* is suspected. The rectum may be only positive post-treatment site. Insert a swab about 2.5cm into the anal canal, just inside the anal ring. Move the swab from side to side and then remove it. No fecal material should be on the swab.
- If specimen is collected for antigen detection use appropriate swab and transport tube for Gen-Probe Pace

Labeling

- Label the specimen with patient information.
- Indicate the suspected diagnosis.
- Indicate the time the specimen was taken and its specific source.

Transport

- Although *N. gonorrhoea* can survive on swabs for up to 6 hours, viability is inevitably lost over time.
- Specimens to be cultured for GC must be cultured immediately and placed in CO₂.
- Ideally, the swab is inoculated directly onto special media at the patient's bedside. Otherwise, place swab in transport medium and deliver it to the laboratory promptly.
- Do not refrigerate the specimen.
- Gen-Probe Pace 2 swabs may be held at room temperature for 24 hours without loss of DNA for detection.

Comment

- Gram stain cannot be used effectively in women to detect *N. gonorrhoeae* in vaginal or cervical specimens. Other organisms that morphologically mimic this agent are present.

Eye Culture

Selection

- Do not use term "eye" for identifying a specimen. Specify what the specimen is, e.g., lid margin sample, conjunctival sample, corneal sample, aqueous or vitreous sample. Specify left or right eye.
- In serious eye infections such as suppurative keratitis or endophthalmitis, the physician and the microbiologist must communicate so that appropriate media and transport systems are made available. For bacteria, chocolate agar is likely to be a good universal medium.
- The method of specimen collection depends on the site of the eye infection. In bilateral conjunctivitis, culture of a specimen from only one eye is necessary.
- For conjunctival specimens, the laboratory ideally needs two swabs from the infected site: one for culture and one for Gram stain. Better Gram stain results may be obtained with scrapings, not swabs, of the lid margin, conjunctiva, or cornea.

Collection Materials

- Sterile Kimura or instrument for scraping
- Sterile calcium alginate swabs, two per package
- Sterile cotton swabs, two per package
- Frosted, etched glass slides
- Microslide holders
- Alcohol wipes
- Preservative-free, unit-dose 0.5% tetracaine
- Pencil or marker for labeling

Collection Methods

Conjunctivitis:

- Obtain specimens before instillation of topical anesthetic. Moisten cotton or alginate swab with broth (unless exudate is present), and scrub swab over inferior tarsal conjunctiva and fornix of infected eye. An additional swab can be taken for Gram staining. Use viral or bacterial transport per request.

Keratitis:

- Do not submit corneal smear or anterior chamber fluid on swab. Take conjunctival specimens with calcium alginate swab, and make single row of C-shaped streaks on chocolate agar. Second swab is used for fungal medium if fungi are suspected. Scrape corneal ulcer with spatula or no. 15 blade scalpel. After anesthesia, scrape cooled spatula over surface of area of suppuration in short, moderately firm strokes in one direction without touching lashes or lids. Use each scraping to inoculate another row of C shapes on chocolate plate or to make a smear. For viral keratitis, conjunctival exudate and scrapings in viral transport are required. Virus is usually shed into tears of cul-de-sac, making conjunctival viral culture.

Endophthalmitis:

- Conjunctival cultures may provide minimal clinically relevant information if used alone. Sampling purulent wound abscess may be helpful, but most useful information may come from aspirate of intraocular fluid from patient in operating room. Obtain anterior chamber and vitreous fluid. Need 1-2ml of vitreous fluid obtained by aspiration or, ideally vitrectomy. Medium should be available at bedside.

Labeling

- Label the specimen with the actual diagnosis, not "eye".
- Label the specimen as being from the right or the left eye.
- Label the specimen with patient information.

Transport

- Many specimens should be plated at the specimen collection site, e.g., the eye clinic. The small amount of material collected tends to dry quickly, and this drying may contribute to a loss of viability of agents.
- Use anaerobic transport where necessary but not for conjunctival specimens.
- Chill the viral transport medium for transport.

Specimen Collection - Group A Streptococcus (throat)

**NOTE: Throat specimens are the only specimen approved for testing.
Collect the specimen using approved swab transport systems**

A. Collection Swabs

- BD or Copan Liquid Amies Medium with Dacron Tip

B. Procedure

- Using an appropriately labeled swab(s), swab the posterior pharynx, including any visible lesions and irritated areas.
- Swab both sides of the throat.
- Return the swab(s) to the transport container.
- Store and transport at room temperature (15-25°C)

COPAN LQ AMIES Transport Swab (Red Cap/Blue Label)



Specimen Collection - Group B Streptococcus (vaginal, perianal swabs)

A. Collection swabs

- BBL or **COPAN LQ Stuart Culture Swab**
- Liquid Stuart **Copan Transystem**
- Liquid Stuart **Copan Venturi System**
- Liquid Stuart **HealthLink Transporter**

NOTE: *This assay includes both the culture of the vaginal/perianal swab specimen in LIM Broth followed by real-time PCR*

B. Procedure

- Insert one **Molecular Testing Swab** into the distal third of the vagina and sample secretions from the mucosa
- Rotate the Molecular Testing Swab for 30 seconds to ensure adequate sampling.
- Carefully withdraw the Molecular Testing Swab and place into appropriately labeled transport container.

- Insert second Molecular Testing Swab approximate 2.5 cm beyond the anal sphincter.
- Gently rotate at least one full turn to sample anal crypts.
- Carefully withdraw the Molecular Testing Swab and place into above labeled transport container.
- Store and transport at room temperature (15-25°C).
- Alternately, the two swabs can be used to sequentially collect first a vaginal specimen and then a perianal specimen.

COPAN LQ AMIES Transport Swab - Double Swab (Red Cap/Blue Label)



COPAN LQ AMIES Transport Swab - Single Swab (White Cap/Blue Label)



HPV (cervical scraping/biopsy)

A. Collection Using [SurePath](#) Preservative Fluid (ThinLayer)

- Collect the cervical sample according to the Pap smear guidelines using the broom-like collection device provided.
- Using the thumb and forefinger of a gloved hand, disconnect the head of the device from the handle and insert the head into the appropriately labeled collection vial. Do not touch head of device. Discard handle
- Tightly cap the vial and shake.
- Store and transport at room temperature (15-25°C).

NOTE: *This collection device is for the PrepStain Pap ThinLayer Smear and HPV molecular testing.*

B. Collection Using [Digene Cervical Sampler](#).

1. Cervical Brushing

NOTE: *Collect Pap smear specimen (separate collection device) before obtaining specimen for HPV DNA testing. Collect the HPV DNA specimen prior to the application of acetic acid or iodine if a colposcopy is being performed. This collection device is for HPV molecular testing only.*

- Collect the Pap cervical sample according to the Pap smear guidelines using the brush-like collection device provided.
- Insert the brush to the bottom of an appropriately labeled collection device.
- Snap off shaft of brush handle at score line. Cap device.
- Store and transport the specimen at room temperature (15-25°C).

2. Cervical Biopsy

- Collect a fresh cervical biopsy specimen up to 5 mm in cross-section by standard methods.
- Immediately place the biopsy specimen into an appropriately labeled collection device.
- Store and transport the specimen at <-20°C.

C. Collection Using [Cytoc PreservCyt Collection \(ThinPrep\)](#)

NOTE: This collection device is for the ThinPrep Pap Smear and HPV molecular testing.

- Collect the cervical sample according to the Pap smear guidelines using the broom-like collection device provided.
- Separate the broom part of the device from the handle by pulling apart, leaving the broom in the appropriately labeled collection device.
- Tightly cap the vial and shake.
- Store and transport the specimen at room temperature (15-25°C).

HPV Testing Supplies:

Sure-Path Liquid PAP Smear Collection Kit



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Thin Prep Liquid PAP Smear Collection Kit



DiGene Cervical Brush/Collection



MRSA ADMISSION SCREEN

A. Collection Swabs

- BBL or **COPAN LQ STUART Transport Swab**(Red Cap/Red Label)w/ Breakaway Tip
- BBL Culture Swab Plus Amies Gel without charcoal
- Copan Venturi Transystem Amies Gel without charcoal

Procedure

- Using an appropriately labeled swab, insert paired swab approximately 1/2 inch into nostril.
- Rotate swab against inside of nostril for 3 seconds.
- Apply light pressure on the outside of the nose while rotating swab.
- Repeat for other nostril with same paired swab.
- Return both swabs to same transport tube and submit to lab at room temperature (2-30°C) is acceptable.

NOTE: A dry swab may be used but must be pre-moistened with sterile physiological (non-bacteriostatic) saline.

COPAN LQ STUART Transport Swab (Red Cap/Red Label) w/ Breakaway Tip



Nasal Specimens

Selection

- The specimen of choice is a swab specimen taken at least 1 cm inside the nares. . Lesions in the nose require samples from the advancing margin of the lesions.

Collection Materials

- Swab and transport medium set
- Nasal speculum (for some patients)

Collection Method

- Use non-bacteriostatic saline to moisten swab.
- Carefully insert the moist swab at least 1 cm into the nares.
- Firmly sample the membrane by rotating the swab and leaving it in place for 10 to 15 seconds.
- Withdraw the swab, insert it into a transport container, and crush the vial of transport medium in the container.

Labeling

- Label the swab container with patient information.
- Indicate whether or not a lesion is present.

Transport

- Transport the specimen to the laboratory as soon as possible.
- Do not refrigerate the specimen.

Comments

- Anterior nares cultures, without an indication of the presence of a lesion, are routinely examined only for *Staphylococcus aureus* and beta-hemolytic streptococci.
- Nasal cultures do not predict the etiologic agent of sinus, middle ear, or lower respiratory tract infection and should not be submitted in lieu of specimens from these sites.
- Anaerobic cultures are not done on nasal specimens.
- Detection of carriage of MRSA may be increased by also sampling another body site, such as the rectum. Two or three consecutively negative samples from these areas usually indicate that the patient is free from carriage. In any case, the presence of MRSA in a patient should not preclude the patient's admission or readmission to a nursing home or hospital.

Rectal and Anal Swab Specimens

Selection (In most cases, the specimens of choice for culture of bacterial agents of diarrhea is portion of the diarrheal stool, not a swab.)

- Rectal swabs are acceptable only for culture of diarrheal pathogens from infants or from patients who are acutely ill with diarrhea.
- Swabs for culture of enteric pathogens must show feces. Anal swabs are not acceptable for culture of bacterial agents of diarrhea.
- Routine culture usually indicates a search for Salmonella, Shigella, and Campylobacter spp. and E. coli O157:H7. If other agents are suspected, consult the laboratory.
- For group B streptococcus isolation, see "General Specimens: General Information" later in this section.

Collection Materials

- Swab
- Transport medium

Collection Method

- Gently insert the swab beyond the anal sphincter, rotate the swab, remove it, and place it into transport medium. The swab should show feces.
- For Neisseria gonorrhoea (GC) cultures, swab the anal crypts just inside the anal ring. Avoid fecal contamination as much as possible.
- Place the GC swab into transport medium immediately, or inoculate a special GC plate at the patient's bedside or at the examination table. Consult the laboratory for the appropriate medium to use.

Labeling

- Label the sample with the patient information.
- Indicate the pathogens sought, especially if GC is suspected.
- Indicate the time of collection on the requisition form.

Transport

- For N. gonorrhoea, do not refrigerate the specimen, but get it to the laboratory within 30 minutes of collection if possible.
- For routine culture, refrigerate the transport medium if a delay in transit of 6 hours or more is anticipated.

Comments

- The specimen of choice for diagnosing the bacterial agent of diarrhea is the diarrheal stool. *N. gonorrhoea* is very fastidious and will die at refrigerator temperatures, without carbon dioxide, or without its proper culture medium.
- If organisms such as *Escherichia coli* O157:H7, *Yersinia* spp., and in some cases *Vibrio* spp. and *Aeromonas* or *Plesiomonas* spp. suspected, notify the laboratory.
- A rectal swab is not recommended for detection of the toxin of *Clostridium difficile*.

Skin and Contiguous Tissue Specimens

(Wound, Abscess, Burn, Exudate)

Selection

- The specimen of choice depends on the extent and character of the infection rather than on the suspected pathogen.
- For most open lesions, remove the superficial flora before collecting a specimen from the advancing margin.
- For dry, encrusted lesions, culture is not recommended unless an exudate is present.
- A closed abscess is the specimen site of choice. Collect exudate and sample of the wall abscess wall.
- For an open abscess, decontaminate the lesion first, as for other open lesions.
- Culture burn wounds only after extensive cleaning and debridement. Biopsy specimens are recommended. Quantitative culture of burn surfaces may or may not be of value.
- The specimen of choice is taken from the advancing margin of the lesion, not just pus. Remove the exudate to get to the interior of the lesion.

Collection Material

- Skin disinfectant
- Sterile swabs - Anaerobic (blue cap w/ Gel) or aerobic (red cap)
- Syringe and needle

Collection Method

- Unruptured abscess. Do not swab. Decontaminate the skin overlying the abscess, and aspirate abscess contents with a syringe. After excision and draining, submit a portion of the abscess wall for culture. Submit the specimen in an anaerobic transport container.
- Open lesions and abscesses. Remove as much of the superficial flora as possible by decontaminating the skin. Remove exudate, and firmly sample the margin of the lesion with a swab. Submit the swab in aerobic transport medium. You can also culture a sample of the exudate aerobically. Do not request anaerobic cultures from open, superficial lesions. Consult with the laboratory.

- Burn wounds. Debride the area, and disinfect the wound. As exudate appears, sample it firmly with a swab. Submit the sample for aerobic culture only. Submit biopsy tissue as the specimen of choice. Surface specimens usually represent only colonization.

Labeling

- Do not label specimen only as "wound" without giving a specific description and the anatomic source.
- Label the specimen with the patient information.
- Note whether the exudate is from an open or closed wound.
- Indicate aerobic or anaerobic culture requests.
- Aerobic only. Submit in aerobic transport medium.
- Superficial-lesion exudates
- Open-wound exudates
- Laceration exudates
- Open-abscess exudates
- Anaerobe and aerobe requests. Submit in blue-capped culturette with gel for anaerobe.
- Surgical aspirates
- Closed-abscess
- Biopsy tissue

Transport

- Transport the specimen to the laboratory quickly.
- Refrigerate the specimen if it is not culture within 1 hour.

Comments

- Skin decontamination is critical to proper culture interpretation.
- The laboratory will Gram stain the specimens. The presence of epithelial cells in the smear indicates surface contamination, and the results culture will be compromised. The presence of leukocytes in the absence of epithelial cells represents an appropriate specimen.
- Do not submit pus only. Pus is not a representative specimen of the lesion. Sample the advancing margin.

Sputum

Selection

- Sputum may not be the specimen of choice for determining the etiologic agent of bacterial pneumonia. Blood specimens, lavage specimens, or transtracheal aspirates may be more accurate.

- Lower respiratory tract secretions from infected patients are confirmed by noting the presence of large numbers of leukocytes in the absence of epithelial cells. Since epithelial cells in the specimen signal gross contamination with the oropharyngeal flora, culture only specimens that represent infection. Careful attention to the instructions given the patient will greatly reduce the number of inappropriate specimens. The first, early-morning specimen is preferred. Pooled specimens are not recommended for culture.
- Handle all lower respiratory tract specimens with the safety precautions necessary for working with *M. tuberculosis*.

Collection Materials

- Sterile, screw-cap sputum collection cup.

Collection Method

- Instruct the patient in the difference between sputum and spit. Explain that a deep cough the first thing in the morning, if practical, is needed to produce a sputum sample.
- Have the patient rinse the mouth with water. For patients with dentures, remove the dentures first.
- Collect the specimen directly into the container.
- Carefully and tightly replace the cap. Be careful not to misthread the lid, because leakage will occur, thus signaling the laboratory to discard the entire container before culture. Check the top to ensure that it is secure.
- Submit the capped tube containing the specimen, and discard the outer package.

Labeling

- Label the specimen with patient information.
- Indicate whether the specimen is for routine, acid-fast bacillus, *Legionella* or fungal culture.

Transport

- Refrigerate the specimen if a delay of >1 to 3 hours is anticipated.
- Transport the specimen to the laboratory quickly.

Comments

- A single, properly collected specimen should be adequate for the diagnosis of bacterial lower respiratory tract disease.
- For the diagnosis of fungal or mycobacterial disease, separately process three consecutive early-morning specimens. Culture of more than five specimens a week for these agents is not indicated. . Anaerobic studies of sputum are not done. . Sputum is not appropriate for the detection of Pneumocystis

Stool or Feces for Culture or Parasitology Studies

Selection

- The specimen of choice is a diarrheal stool (the acute stage of illness.)
- A rectal swab for bacterial culture must show feces. Generally, swabs are recommended only for infants.
- For bacterial pathogens, collect and submit three specimens, one each day for three days.

For parasite examination, three specimens collected every other day or every third day should be adequate. A single stool specimen may not exclude bacterial or parasite pathogens as a cause of diarrhea.

- To rule out the carrier state for some organisms, three consecutive negative specimens are often needed. Specimens for parasite examination collected too soon after administration of barium, oil, magnesium, or crystalline compounds are unsatisfactory. Delay specimen collection a minimum of 5 days after administration of these agents.

Collection Materials

- Commode collection system in which a plastic collection device fits over the rim of the toilet seat are available. Alternatively, a clean, waxed cardboard cup with secure lid or some other similar container (when the specimen is for bacterial culture and immediate parasitology examination) can be used. The smaller the container, the more difficult it is for the patient to provide an appropriate specimen.
- Swab (for acutely ill patients or when stool specimen is not available).
- Parasitology transport pack (one vial of SAF) or equivalent.

Collection Method

- Instruct patients who can to excrete directly into the cup or collection device: Never take a specimen from the water in a toilet. Do not allow urine to contaminate the specimen. Replace the lid tightly, and refrigerate specimen.
- Alternatively, collect feces from a sterile bedpan, and place 10 to 20 g into container.

- For parasite studies, use either method described above and then bring the specimen directly to the laboratory while the specimen is still warm. If a delay is necessary, place about 0.5 to 1 teaspoon of specimen into each fixative provided.

Labeling

- Label the specimen with patient information.
- Indicate the type of studies required: routine culture, ovum and parasite, special studies etc.
- Indicate the time and date of collection.
- Indicate any special patient history (travel, other ill family members, etc.).
- Label as a specimen in a series for a single patient if appropriate (e.g., 1 of 3, 2 of 3, 3 of 3).

Transport

- If the specimen is not transported immediately for bacterial culture, refrigerate it.
- If the specimen is to be submitted for *C. difficile* study and a >48 hour delay is anticipated, freeze the specimen or submit it quickly at 4°C.
- Submit fresh specimens for parasite studies as quickly as possible. Preserved specimens need not be rushed to the laboratory.
- For bacterial pathogens, use a vial with Cary-Blair transport medium if a 2 to 3 hour delay is anticipated.

Comments

The laboratory must be notified if bacterial other than *Salmonella*, *Shigella*, *Campylobacter* spp. or *E. coli* 0157:H7 are suspected as the cause of diarrhea. Isolation of *Vibrio*, *Yersinia*, or *Aeromonas* spp. requires special procedures in the laboratory and usually more expense.

- Susceptibility studies are not routinely done on *Campylobacter* isolates or *E. coli* 0157:H7.
- Anaerobic studies are not done on feces.
- Transport bile, colostomy, and ileostomy specimens in the same manner as other fecal specimens.
- Small-bowel aspirates can be tested for anaerobes. *Bacteroides* and *Bifidobacterium* spp. can colonize the small bowel and cause a malabsorption syndrome in the presence of an obstruction.
- We offer an initial parasite screen for *Giardia lamblia* and *Cryptosporidium* spp. rather than a complete ovum and parasite microscopic examination. Modern enzyme immunoassay or fluorescent detection methods make it unnecessary, in most cases, to provide three fecal samples. Many studies have shown that one sample is usually enough to detect antigens.
- The more immunosuppressed a patient, the more likely it is that some parasites may disseminate to other body sites, requiring additional clinical specimens to be sent to the laboratory.

Specimen Collection - Stool ParaPak

Please follow instructions carefully as the quality of your specimen is critical to the accuracy of your test results.

*NOTE: DO NOT use a laxative before collecting a stool specimen. Use the special stool transport systems:

- Ova and Parasites (**ParaPAK** Pink and Blue - Formalin, 10% and PVA fixative)
 - Stool Culture (ParaPAK Orange - Enteric Transport C&S)
 - Available for pickup at Synergy Laboratory Patient Service Centers or MSHA Facilities.
 - Clients may order by contacting Support Services.
1. Collect the stool in a clean wide mouthed container. Urine or water must not contaminate the sample. Urinate before collecting the stool specimen, if necessary.
 2. Remove the vials from inside the plastic bag.
 3. Open the vials carefully. Using the collection spoon attached to the vial cap, add enough stool until the liquid reaches the FILL line on the vial label, (approximately the size of a walnut).

DO NOT OVERFILL the vials.

NOTE: Collect stool from areas that look bloody or have mucous.

4. Tighten the cap so the specimen cannot leak out and shake the vial until the mixture appears well mixed.
5. Label the containers with the patient name, date and time of collection.

*Store at room temperature until delivered to the laboratory.

*Return the specimen to a Synergy Laboratory Affiliate location within 24 hours of collection.

Clostridium difficile testing:

1. Collect a fresh sample as above and place the sample in a **specimen cup** with no preservative.
2. Deliver to the laboratory within a few hours or refrigerate.
3. Return to a Synergy Laboratory Affiliate location within 24 hours of specimen collection.

Molecular Testing Collection Supplies

*ParaPAK Pink and Blue
(10% Formalin and PVA Solutions)*



*ParaPAK Orange (Enteric Specimen
Collection)*



Sterile Collection Container



Throat Specimens

Selection

- Success with culture or with direct antigen detection depends on firmly and completely sampling an area of the inflamed throat.
- Using a tongue blade to hold the tongue down, look at the back of the throat and the tonsillar area for localized areas of inflammation and exudate.
- These areas are the most productive for producing cultures of the etiologic agents of acute pharyngitis.

Collection Materials

- Dacron or calcium alginate swab and transport medium.
- Tongue blade

Collection Method

- Carefully but firmly rub the swab over several areas of exudate or over the tonsils and posterior pharynx.
- Do not touch the cheeks, teeth, or gums with the swab as you withdraw it from the mouth.
- Insert the swab back into its packet, and crush the transport medium vial in the transport container.

Labeling

- Label the swab container with patient identification data, including the time of collection.
- Note any antimicrobial agents currently being taken by the patient.
- Note whether the specimen is for culture or direct antigen detection. Indicated whether it is for throat culture or a streptococcus screen.
- Indicate the suspected pathogen if other than streptococci, e.g., *N. gonorrhoea*.

Transport

- Transport the swab to the laboratory as soon as possible.
- If transport is to be delayed beyond 1 hour, refrigerate the swab.

Comments

- A streptococcus culture will test for and report the presence or absence of beta-hemolytic streptococci, including group A, group C, and G. A throat culture must be ordered if the physician is looking for other potential respiratory pathogens, including *Streptococcus pneumoniae*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, or *Neisseria gonorrhoeae*.
- Beta-hemolytic streptococci are routinely identified and reported. They are the primary cause of acute bacterial pharyngitis and need not be tested for susceptibility to antimicrobial agents.
- *Haemophilus* spp. may be reported in pediatric patients when requested, although they are part of normal flora in adults and children.

- Throat culture for N.gonorrhoeae is available on request, but the laboratory must be alerted to the request.
- Susceptibility testing is not routinely done on any isolate from the throat, although a screen for methicillin-resistant S. aureus (MRSA) may be requested if the carrier state for MRSA is suspected.

Urine Collection

Random Urine

Supply the patient with clear verbal and written instructions as follows.

Explain, "We need a good urine specimen to diagnose your infection. It is important that you understand this procedure so that you will not contaminate the specimen with other germs. "

Check that the top of the cup is secure and not misthreaded. A leaking container is dangerous to patients and personnel alike. Refrigerate the specimen if it is not brought immediately (within 30 minutes) to laboratory or if it is transported in urinalysis tube without preservatives. **DO NOT transport urine to the laboratory in the screw top cup.** Use a Direct Draw Adaptor or Urine Collection Straw to fill evacuated urinalysis tube. This tube can easily be distinguished from the blood tubes by the **BLUE** label and the **CONICAL** shaped bottom.

Clean Catch Instructions for Females:

1. Sit comfortably on the toilet, and swing one knee to the side as far as you can.
2. Spread yourself with one hand, and hold yourself spread while you clean yourself and then collect the specimen.
3. Wash - be sure to wash and rinse well before you collect the specimen. Using the cleaning materials supplied, wipe your vaginal area as carefully as you can from the front to the back between the folds of the skin.
4. Rinse. After washing with each soap pad, rinse with a water-moistened pad with the same front-to-back motion. Use each pad only once, and throw it away.
5. Hold the cup with your fingers on the outside; do not touch the rim.
6. First, pass a small amount of urine into the toilet, and then pass enough urine into the cup to fill it half full. Place the lid on the cup carefully and tightly, or ask the nurse to do it for you.

Clean Catch Instructions for Males

1. Retract the foreskin (if uncircumcised), and clean the glans (head of the penis).
2. Follow steps 3 to 6 above for cleaning yourself and collecting the urine.
3. Labeling
 - Label the specimen with patient information.
 - Indicate on the request form whether or not the patient is symptomatic and taking antibiotics.

Comments

- Culture urine from pediatric bags immediately to minimize interference by contaminants.

- Routine urine samples are unacceptable for anaerobic culture.
- Boric acid tubes with Vacutainer collection devices are preferred for transport of urine since urines held in boric acid are stable at room temperature for 24 hours. However, no other testing will be possible on the Boric Acid sample. If additional testing is required (i.e. UA, Urine Electrolytes, etc.), a second sample must be submitted.

Catheter Collection Method

- If necessary, clamp the catheter tubing to collect urine in the tube, but do not allow the clamp to remain for more than 30 minutes.
- Clean the sampling port (or tube site if a port is unavailable) with alcohol swabs.
- Insert the needle into the tubing port, and withdraw urine onto the syringe.
- Transfer the urine to a sterile cup or boric acid transport tube.

Labeling

- Label the specimen with patient information (Name, numeric identifier, Collection Date/Time & Initials of person collecting specimen).
- Indicate on the request form that the specimen was taken from an indwelling catheter. Transport
- Refrigerate the urine if it is not delivered to the laboratory within 30 minutes of collection.

Comments

- Do not disconnect the catheter from the catheter bag to collect the specimen, never submit bag contents for culture.
- Patients with indwelling catheters will probably be colonized after 48 to 72 hours, often with multiple isolates.
- The laboratory must know whether the urine is collected by any method that might introduce contamination: for example, collection at home in an unconventional container, unknown collection method (e.g.. form nursing home, etc.).

24 hr Urine Collection

A 24 hour urine jug is supplied for those assays that require a 24 hour urine collection. Please note that some 24hr urine tests may require a preservative. Follow guidelines required for each test accordingly to ensure proper specimen integrity. An instruction sheet will be provided with the jug. An aliquot bottle is also dispensed with the urine jug to our client offices. If the patient returns the 24 hr. urine jug to the laboratory, the laboratory staff will measure and aliquot the urine. If the 24 hr urine jug is returned to the physician office site, the following must be performed:

1. Note the volume on the original collection container.
2. Mix well by inversion and pour an aliquot into the small plastic container provided.
3. Label with all pertinent information, eg. Total volume, collection start and end date & time, test(s) ordered, patient name, ID (medical record number or social security number)
4. Place labeled aliquot bottle into specimen transport bag with requisition.

Do not send the entire 24 hr urine jug. The couriers cannot legally transport those sample container

Suprapubic Aspirate for Urine Cultures

Selection

- This technique avoids contamination of urine with urethral or perineal bacteria.
- The method is required for diagnosing anaerobic urinary tract infections and is most frequently used for pediatric patients, with spinal cord injury, and patients for whom a definitive culture has not been obtained.

Collection Materials

- Supplies for skin decontamination
- local anesthetic
- 22-gauge needle and syringe
- Sterile urine container or boric acid tube for culture

Collection Method

- Decontamination and anesthetize the skin.
- Introduce the needle into the full bladder at the midline between the symphysis pubis and the umbilicus, 2cm above the symphysis.
- Aspirate about 20ml of urine from the bladder.
- Transfer the urine aseptically into a sterile screw-cap container or boric acid tube for transport to the laboratory.

Labeling

- Label the specimen with patient information
- Indicate on the request form that this is a urine aspirate.
- Indicate whether or not anaerobic studies are required.
- Indicate the time of collection.

Transport

- If the specimen is not transported to the laboratory within 30 minutes of collection, refrigerate it.

Comments

- Anaerobic studies are done only on request.
- This procedure can be used on pediatric patients to confirm the positive results obtained from a strapped-on bag device.

Urethral and Penile Specimens

Selection

- The urethral is the male genital site most commonly cultured.

- Remove the external skin flora of the urethral meatus as in preparation for a urine specimen.
- Material from a site about 2cm inside the urethra (by swab) or expressed pus is the specimen of choice.

Collection Materials

- Urethrogenital swab
- Transport medium
- Slide for stained smear

Collection Method

- Express exudate from the urethra, and collect the exudate on a swab. Place the swab into transport medium.
- Collect additional exudate on a swab, and use this swab to prepare a slide for staining. Roll the swab over 2 to 3cm of the slide surface, and label the same side.
- If exudate is unavailable, insert an urethrogenital swab about 2cm into the urethra, gently rotate the swab, and remove it.
- For culture transport to the lab immediately or inoculate the specimen onto special medium as soon as possible, and place the specimen into a CO₂ atmosphere at 35°C. If two swabs are available, submit one for culture and prepare a smear with the other.
- For antigen detection of *Neisseria gonorrhoea* and *Chlamydia trachomatis*.
- Collect exudate using Gen-Probe swab for Pace 2 assay.

Labeling

- Label the specimen with patient information.
- Indicate the time of collection.
- Indicate the suspected diagnosis.

Transport

- Do not refrigerate the specimen. (VERY important for routine culture)
- Transport the specimen to the laboratory immediately.

Comments

- Diagnosis of gonorrhoea in males can often be confirmed by Gram stain of urethral exudate. For female, confirmation by Gram Stain of GC in vaginal or cervical secretions cannot be done because some nonpathogenic species in the vaginal may resemble the diplococcal morphology of *N. gonorrhoea*.
- *N. gonorrhoea* is nutritionally fastidious and environmentally fragile and cannot tolerate cold temperatures or lack of CO₂.
- Along with the test for *N. gonorrhoea*, consider a request for Chlamydia detection, because this agent is often found in patients with urethritis.
- The choice of a swab type is critical. Check the package insert to determine whether cotton tipped, calcium alginate, or Dacron-tipped swabs should be used with the selected procedure.

