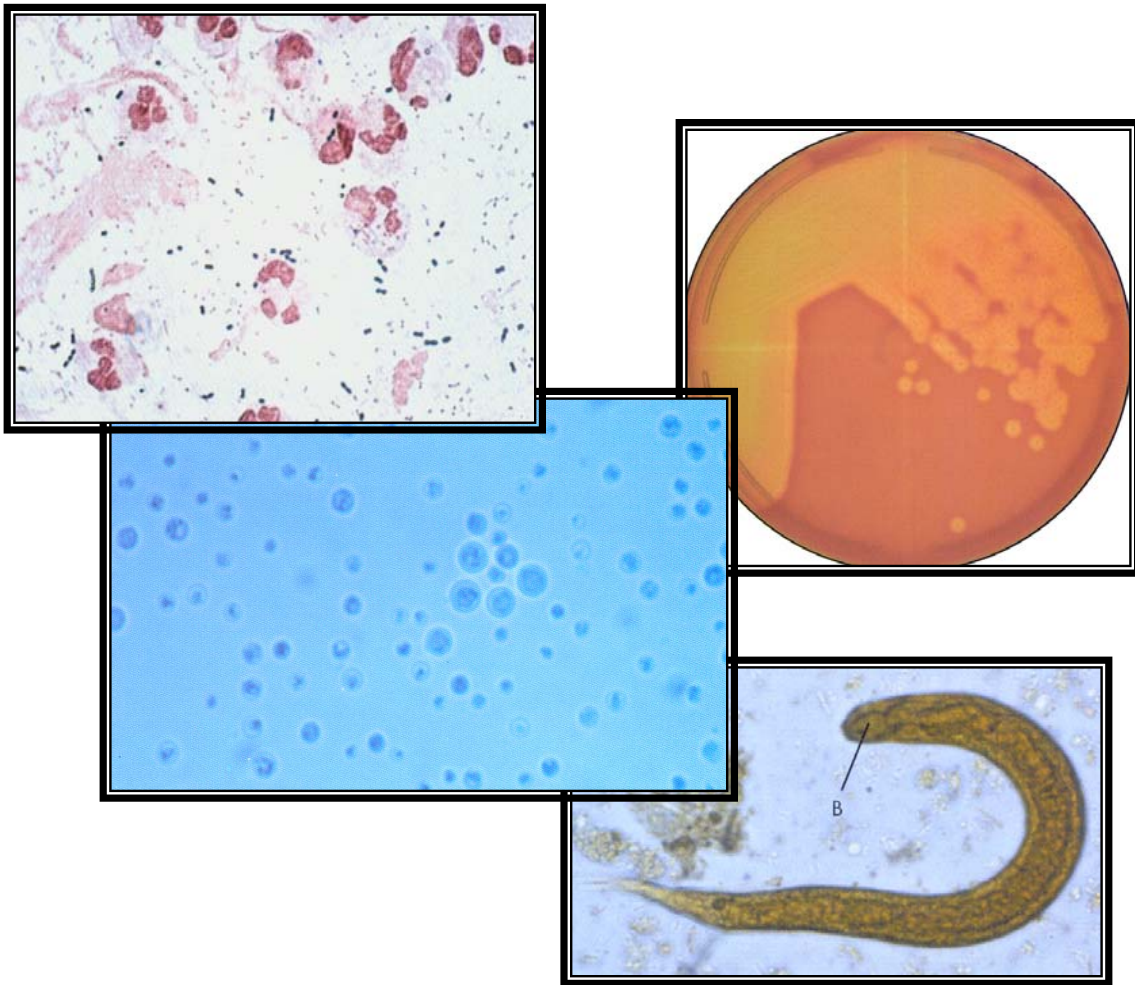


Clinical Microbiology

A Specimen Management Guide, 2nd Edition (2011)



Medical Staff and Nursing Version

A Product of the San Joaquin General Hospital Laboratory, Department of Microbiology

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Introduction

Purpose

The effectiveness of a laboratory is relied upon the importance of specimen management. This involves specimens that are appropriately selected, collected, labeled and transported to the laboratory. If a specimen is not collected or managed properly, the laboratory can do very little or nothing in the patient care and/or further investigations. The proper management of patient specimen is critical in achieving accurate diagnosis. This accuracy directly affects patient care, treatment and length of stay. It also affects the hospitals infection control, overall hospital cost, laboratory costs and influences laboratory efficiency. (1,2)

It is the responsibility of the laboratory, to communicate its needs, as well as understand the needs of the physicians involved. Communication between laboratorians and the medical staff is imperative for accurate and efficient care that the patient receives. At times, physicians and microbiologist need to have constructive interaction with one another. This may involve asking question to clarify test requests, suggestion of equal but more cost effective alternatives, requesting additional information, or even rejecting a specimen due to improper care of the specimen prior to its arrival at the laboratory. (1)

Just as there are standards for patient care that the clinician must follow, there also are standards for laboratory practice that the laboratorians must follow. Challenges are met every day as to the integrity of a patient specimen. At times, patient specimens should not be processed due to poor management of the specimen. This manual is designed to avoid improper management of patient specimen by assisting all the members of the health care team that are involved in the process of specimen selection, collection, labeling and transporting to the laboratory. This will ensure the likelihood of recovering viable organisms for culture. Hence, the “Clinical Microbiology, A Specimen Management Guide” was created. (1)

Labeling

Labeling of specimens is of high importance. All specimens should be properly labeled to ensure that an accurate diagnosis is being reported on the correct patient. Also of importance is that the correct tests are being applied to the correct container or specimen type. When labeling, make sure the specimen container belongs to the patient in question. The label should include the patients name, medical record number or date of birth, date of collection, time of collection, **First initials and complete Last name** of collector and anatomical site when applicable (i.e. left arm, right foot, pleural fluid, ascites fluid, liver tissue, skin tissue, etc., etc...)(**See Figure A:** example label). The worst scenario would involve diagnosis given on a patient where medical treatment is carried out due to improper labeling, which may result in patient complications or

even death. So it is very important to take your time in correctly labeling patient specimen to ensure correct diagnosis. (1,2)



Figure A

Safety

Biosafety is of utmost importance in the laboratory. It is a concern to the laboratorian and to other health care workers in a health care system. To keep abreast of new methods and techniques that insure a safe working environment, please refer to your hospital safety manual. Related policies for safety in specimen management include the following:

- 1) All specimen collection procedures must be performed while wearing gloves, a laboratory coat, and, where appropriate, masks and /or goggles
- 2) All primary specimen containers should be leak-proof and should be transported within a sealable, leak-proof plastic bag having a separate compartment for paperwork
- 3) Never transport syringes with needles to the laboratory. Instead, the contents should be transferred to a sterile tube or the needle should be removed with a protective device, and the syringe should be recapped and placed in a sealable, leak-proof plastic bag
- 4) Do not transport leaking specimen containers to the laboratory or process them. (2)

Transport Policy

All specimens must be promptly transported to the laboratory, preferably within 1 hour of collection. If specimens are not processed immediately, store specimens for bacterial culture in refrigerator for not more than 24 hours. Optimal transport of clinical specimens, including

anaerobic cultures, depends primarily on the volume of material obtained. Submit small amounts within 15-30 minutes of collection; biopsy tissue can be maintained for up to 20-24 hours, if stored at 25 °C in an anaerobic transport system. Environmentally sensitive organisms including *Shigella* spp., *N. gonorrhoeae*, *Neisseria meningitidis*, and *Haemophilus influenzae*, which are sensitive to cold temperatures, should be process immediately. Never refrigerate spinal fluid, genital, eye, or internal ear specimens or specimens suspected of containing these agents. (1)

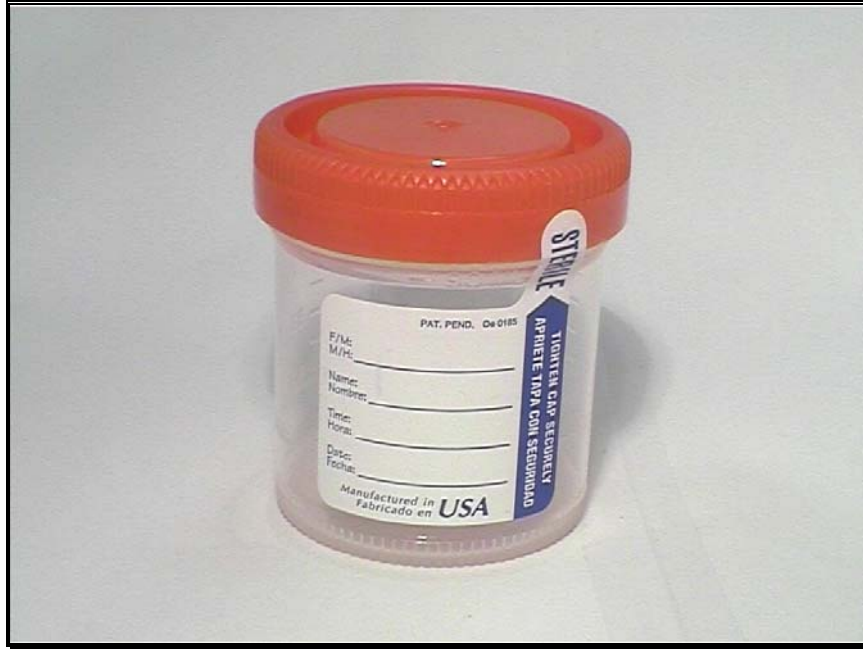
Summary of Microbiology Tests

<u>Test description</u>	<u>OC Mnemonic</u>	<u>Page</u>
AFB Culture (<i>includes AFB smear</i>)	AFBC	1, 22, 25
AFB Culture Blood/MAC/MAI	AFBL	16
Biopsy/Tissue Culture Panel (<i>includes Gram stain, aerobic and anaerobic culture</i>)	CXB	29
Blood Culture Set #1	BCX1	5
Blood Culture Set #2	BCX2	5
Blood Culture Set #3	BCX3	5
Blood Culture Pediatrics	BCXP	6
Body Fluid Culture: (<i>includes Gram stain, aerobic and anaerobic culture</i>)		
Abdominal	CXBF	28
Amniotic	"	"
Ascites	"	"
Bile	"	"
Joint	"	"
Paracentesis	"	"
Pericardial	"	"
Peritoneal	"	"
Pleural	"	"
Synovial	"	"
Thoracentesis	"	"
Catheter Tip Culture	CXCAT	1
Cerebrospinal Fluid (CSF) Culture (<i>includes Gram stain</i>)	CXCSF	22
Chlamydia:		
Culture (Non-sexual assault)	CHLCX	10
Sexual Investigations (Sexual assault)	CHLSI	10
/ GC by DNA probe	CHLGC	13
Clostridium Difficile Toxin A & B	CDIFF	1
Cryptococcus neoformans Antigen	CRYPA	22
Diphtheria Culture	CXDIP	8
Ear Culture Panel (<i>include Gram stain</i>)	CXEAR	8
Eye Culture Panel (<i>include Gram stain</i>)	CXEYE	8
Fungal Culture (Routine)	CXF	1, 22, 25
Fungal Blood Culture	FUNBL	7
Fungal Skin/Hair Culture	CXFS	1
Genital Culture Panel (<i>includes Gram stain</i>)	CXGEN	8
Genital Strep B Culture	CXSTB	8
GC Culture Panel (<i>includes Gram stain</i>)	CXGCP	8
GC / Chlamydia by DNA probe	CHDNA	13
Helicobacter Pylori Stool Antigen	HPSA	1
Herpes Culture	CXHER	10
India Ink for Cryptococcus neoformans	IINK	22
KOH Prep	KOH	1, 8, 22, 25
Legionella Culture	CXLEG	1, 22, 25
MRSA Screen Culture	MRSA	8
Mycoplasma/Ureaplasma Culture	CXMYC	11
Ova & Parasite Panel (Routine O&P Screen and Giardia EIA)	OP	17

<u>Test description</u>	<u>OC Mnemonic</u>	<u>Page</u>
Ova & Parasite Extended Profile: (Includes Ova & Parasite Panel, Cyclospora, Cryptosporidium) Isospora, Microsporidia and Sarcocystis)	OPEP	18
Pertussis Culture	CXPER	25
Pinworm (Enterobius vermicularis)	PINWM	24
Pneumocystis carinii DFA	PCP	25
Respiratory:		
Sputum Cough Screen	SPSCR	1
Sputum Induced	CXRES	25
Respiratory Syncytial Virus (RSV) EIA	RSV	1, 25
Rotavirus EIA	ROTAV	1
Stool-Feces Culture	CXSTL	1, 19
Stool WBC	STWBC	1
Streptococcus A (Rapid Test)	STRPA	8
Throat Culture	CXTH	8
Urine Culture		
Catheter	CXUA	1, 20
Clean Catch	"	1, 20
Midstream	"	1, 20
Suprapubic	"	1, 20
Vancomycin Resistant Enterococci (VRE) Screen	VRE	1, 8
Viral Culture:		
Blood / Bone Marrow	CXVIR	12
Cerebrospinal Fluid (CSF)	"	22
Stool	"	1
Urine	"	1
All other specimen types	"	10
Wet Mount	WETMT	19
Wound Culture:		
Deep Wound Panel (includes Gram stain, aerobic and anaerobic culture)	CXWDD	14, 27
Superficial Wound Panel (includes Gram stain and aerobic culture)	CXWDS	8

This list is not conclusive. If a particular test is desired and is not included in this guide, please call one of the following departments for specimen selection, collection and management requirements:

- | | |
|-------------------------------|------------------------------|
| 1) Sendouts | (209) 468-6961 |
| 2) Specimen processing | (209) 468-7059 |
| 3) Microbiology | (209) 468-6065 / 6064 |



Sterile specimen cup

Please label specimen container before giving it to the patient.

Intended use:

Respiratory (sputum cough screen,) Fungal Cultures (sputum, BAL, TBbx, body fluids, hair, skin, eye fluids or aspirates), AFB sputum, AFB urine, AFB tissues or body fluids, AFB Stool, AFB bronchoalveolar lavage and bronchial washings, Stool cultures, Stool WBC, *H. pylori* Stool Ag, *C. difficile* Toxin A & B, Rotavirus EIA, Routine Viral Cultures (Stool and Urine), Catheter Tip, Body Fluids (abdominal, amniotic, ascites, bile, joint, paracentesis, pericardial, peritoneal, pleural, synovial, thoracentesis), Routine Urine Cultures, Culture from tissue, *Legionella spp.*

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Acid-Fast Bacilli (AFB) Sputum	5-10 ml	ASAP or ≤ 1 hour	25 °C	Specimen is stable at 4 °C. (2)
Urine	40 ml non-preserved	ASAP or ≤ 1 hour	25 °C	Specimen is stable at 4 °C. (2)
Stool	≥ 1 g non-preserved	ASAP or ≤ 1 hour	25 °C	Specimen is stable at 4 °C. (2)
Bronchoalveolar Lavage Fluids and Bronchial Washings	≥ 5 ml	ASAP or ≤ 1 hour	25 °C	Specimen is stable at 4 °C. (2)
Gastric Lavage Fluids	5-10 ml	ASAP or ≤ 15 minutes neutralize pH within one hour of collection	25 °C	Specimen is stable at 4 °C. To neutralize pH, take a 50 ml conical, add specimen and fill with phosphate buffer, centrifuge 20 minutes, decant supernatant and re-suspend sediment with 5 ml phosphate buffer. Process performed by laboratory personnel. (2)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Acid-Fast Bacilli (AFB)(Cont.)				
Body Fluids (CSF, Pleural, peritoneal, And pericardial)	≥ 2 ml	ASAP or ≤1 hour	25 °C	Specimen is stable at 4 °C. (2)
Tissues (Lymph node, skin, other biopsy material)	≥ 1 g	ASAP or ≤1 hour	25 °C	Specimen is stable at 4 °C. (2)
Catheter tip i.v.	5 cm of distal tip	ASAP or ≤ 15 minutes	25 °C	Acceptable i.v. catheters for semiquantitative culture (Maki method): central, CVP, Hickman, Broviac, peripheral, arterial, umbilical, hyperalimentation, Swan-Ganz. (2)
Feces				
Routine culture	≥ 2 g Un-preserved	ASAP or ≤1 hour	25 °C	Do not perform routine stool cultures for patients whose length of stay was >3 days and the admitting diagnosis was not gastroenteritis. Culture and toxin tests for <i>C. difficile</i> should be considered for these patients. (2)
Stool WBC	≥ 2 g Un-preserved	ASAP or ≤1 hour	25 °C	Specimen is stable at 4 °C. (2)
<i>C. difficile</i> toxin A & B	≥ 2 g Un-preserved	ASAP or ≤1 hour	25 °C	Specimen is stable at 4 °C. (2)
<i>H. pylori</i> stool antigen	≥ 2 g Un-preserved	ASAP or ≤1 hour	25 °C	Specimen is stable at 4 °C. Preferred test, more diagnostic than antibody titer. (2)
Routine Viral Cultures	≥ 2 g Un-preserved	ASAP or ≤1 hour	25 °C	Specimen is stable at 4 °C. (2) Specimen is a sendout to Public Health Department.
Rotavirus EIA	≥ 2 g Un-preserved	ASAP or ≤1 hour	25 °C	Specimen is stable at 4 °C. (2)
Fluids				
Abdominal, amniotic, ascites, bile, eye (1), joint, paracentesis, pericardial, peritoneal, pleural, synovial, thoracentesis	≥ 1 ml for bacteria; ≥ 10 ml for fungi; ≥ 10 ml for mycobacteria	ASAP or ≤ 15 minutes	25 °C	Pericardial fluid and fluids for fungal cultures ≤ 24 hours at 4 °C. Amniotic and culdocentesis fluids should be transported in an anaerobic system and need not be centrifuged prior to Gram staining. Other fluids are best examined by Gram staining of a cytocentrifuged preparation. (1, 2) borders of lesions. Note any antifungal therapy taken recently (1, 2)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Hairs Dermatophytosis	approx. 10 hairs	ASAP or ≤1 hour	25 °C	Collect scalp scales, if present, along with scrapings of active borders of lesions.
Legionella spp. Urine	≥ 2 ml	ASAP or ≤ 1 hour	4 °C or Frozen	Specimen should be first-morning void. Test is a sendout to a Reference Laboratory. (4)
Respiratory	≥ 1 ml	ASAP or ≤1 hour	25 °C	Specimen collection via sputum induced, tracheal aspirate fluid, BAL, trans-bronchial biopsy (TBBx), bronchial brushing, pleural fluid, tissue from lung biopsy. Tests is a sendout to a Reference Laboratory. (2,4)
Nails Dermatophytosis	enough scrapings to cover the head of a thumbtack.	ASAP or ≤1 hour	25 °C	Refrigerate if specimen is not cultured within 2 hours. (3, pg. 264) (1, 2)
Respiratory, lower Sputum, expectorate (cough) screen Bacterial	≥ 1 ml	ASAP or ≤1 hour	25 °C Do not refrigerate	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 100x field. (1)
Fungal	3-5 ml	ASAP or ≤1 hour	25 °C	Refrigerate if specimen is not cultured within 2 hours. (3, pg. 264) (1, 2)
Skin Dermatophytosis	enough scrapings to cover the head of a thumbtack.	ASAP or ≤1 hour	25 °C	Refrigerate if specimen is not cultured within 2 hours. (3, pg. 264) (1, 2)
Tissue Routine cultures and fungal cultures (TBBx or open lung biopsy) (1, 2)	≥ 0.5 g, add sterile saline to sample, to prevent drying.	ASAP or ≤ 15 minutes	25 °C; refrigerate only if fungal specimen is not cultured within 2 hours (3, pg. 264)	Always submit as much Tissue as possible Never submit a swab that has simply been rubbed over the surface. For quantitative study, a 2 x 1 cm sample is appropriate, i.e., about 500 mg Some legionellae may be Inhibited by saline. (2)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Urine				
Indwelling catheter	≥ 1 ml	ASAP or ≤1 hour	25 °C	Non-preserve specimen. (2)
Midstream	≥ 1 ml	ASAP or ≤1 hour	25 °C	Non-preserve specimen. (2)
Straight catheter	≥ 1 ml	ASAP or ≤1 hour	25 °C	Non-preserve specimen. (2)
Suprapubic needle aspiration	≥ 1 ml	ASAP or ≤1 hour	25 °C	Non-preserve specimen. (2)
Viruses	≥1 ml	ASAP or ≤ 15 minutes	4 °C	Best specimen is at least 10 ml of clean-voided, first-morning urine. Frequently isolated: CMV, HSV, mumps virus Less frequently isolated: polyomavirus (JC virus), Rubella virus. (2) Test is a sendout to Public Health Department.



**BacTAlert Adult Blood Culture
(Aerobic and Anaerobic)**

Intended use:
Blood specimen for bacteria and Bone marrow aspirate

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Blood Peripheral Bacterial	10-20 ml	ASAP or ≤ 1 hour	25 °C	Acute sepsis: 2-3 sets from separate site, all within 10 min Endocarditis acute: 3 sets from 3 separate sites, over 1-2 hours Endocarditis subacute: 3 sets from 3 separate sites, take ≥ 1 hour apart; if negative at 24 hours, obtain 2-3 more sets. Fever of unknown origin: 2-3 sets from separate sites ≥ 1 hour apart; if negative at 24 hours obtain 2-3 more sets. (2)
Bone marrow	≥ 1 mL (3)	ASAP or ≤ 1 hour	25 °C	Small amount of bone marrow aspirate may be inoculated directly onto culture media. (2)



BacTAlert Pediatric Blood Culture (Aerobic and Anaerobic)

Intended use:

Blood specimen for bacteria and Bone marrow aspirate

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Blood Peripheral Bacterial	1-10 ml	ASAP or ≤ 1 hour	25 °C	Acute sepsis: 2-3 sets from separate site, all within 10 min Endocarditis acute: 3 sets from 3 separate sites, over 1-2 hours Endocarditis subacute: 3 sets from 3 separate sites, take ≥ 1 hour apart; if negative at 24 hours, obtain 2-3 more sets. Fever of unknown origin: 2-3 sets from separate sites ≥ 1 hour apart; if negative at 24 hours obtain 2-3 more sets. (2)
Bone marrow	≥ 1 ml (3)	ASAP or ≤ 1 hour	25 °C	Small amount of bone marrow aspirate may be inoculated directly onto culture media. (2)



Wampole Isolator 10 Fungi / Yeast Blood Culture

Intended use:
Blood specimen Fungi / Yeast

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Blood Peripheral, Fungi / Yeast	1-10 ml	ASAP or ≤ 1 hour	25 °C	
Bone marrow	≥ 1 ml (3)	ASAP or ≤ 1 hour	25 °C	Small amount of bone marrow aspirate may be inoculated directly onto culture media. (2)



CultureSwab (Modified Stuart's)

Intended use:

Cultures for Ear, Eye, Genital Strept B, Strept A, Throat, MRSA Screen (axilla, nasal, groin), VRE, Deep Wound Aerobe, Superficial Wound Aerobe, Genital Culture Panel, *C. diphtheriae* culture *Neisseria gonorrhoeae* (GC)(Best if plated immediately on to Modified Thayer-Martin (MTM) media or sent as an aspirate)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
<i>C. diphtheria</i>	Swab	ASAP or ≤ 1 hour	25 °C	Swab specimen preferred, but can be collected as nasopharyngeal aspirates, washings. (2) Send out to Focus Technologies, Inc.
Ear				
inner	Swab or ≥ 1 ml in sterile screw-cap tube	ASAP or ≤ 1 hour	25 °C	Throat or nasopharyngeal cultures are not predictive of agents responsible for otitis media and should not be submitted for that purpose. (2)
outer	swab	ASAP or ≤ 1 hour	25 °C	For otitis externa, vigorous swabbing is required since surface swabbing may miss streptococcal cellulitis. (2)
Eye				
Conjunctiva	Swab or direct culture inoculation of : BAP and CHOC	plates: ASAP or ≤ 15 minutes swab: ASAP or ≤ 1 hour	25 °C	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. (2)

Specimen type	Device/ volume	Transport time	Transport temp. (°C)	Comments
Eye (Cont.)				
Corneal scrapings	Direct culture Inoculation: BHI with blood, CHOC, and inhibitory mold agar	ASAP or ≤15 minutes	25 °C	It is recommended that swabs for culture be taken prior to anesthetic application, whereas corneal scraping can be obtained afterwards. (2)
Fluid or aspirates	Sterile screw-cap tube or direct inoculation of small amount of fluid onto media.	ASAP or ≤15 minutes	25 °C	Include fungal media. Anesthetics may be inhibitory to some etiological agents. (2)
Genital Culture Panel				
Cervix, Urethral, Vaginal, Lesions, Prostate (male)	Swab Swab or sterile tube	ASAP or ≤1 hour	25 °C	Indicate collection site. (2) More relevant results may occur by adding urine cultures on specimens obtained before and after massage to indicate urethral and bladder organisms. (2) Ejaculate may also be cultured. (2)
Genital Strep B	Swab	ASAP or ≤1 hour	25 °C	Indicate collection site. (2)
MRSA				
Axilla	Swab	ASAP or ≤1 hour	25 °C	Indicate collection site. (2)
Groin	Swab	ASAP or ≤1 hour	25 °C	Indicate collection site. (2)
Nasal	Swab	ASAP or ≤1 hour	25 °C	Indicate collection site. (2)
N. gonorrhoeae (GC)	Non-cotton swabs or ≥ 1 ml aspirate or direct inoculation of MTM (Modified Thayer-Martin)	ASAP or ≤15 minutes	25 °C	Indicate collection site. Neisseria species are sensitive to cold conditions and to cotton tipped swabs (contains fatty acids). (2)
Strep A	Swab	ASAP or ≤1 hour	25 °C	Indicate collection site. (2)
Throat	Swab	ASAP or ≤1 hour	25 °C	(2)
VRE				
Rectal	Swab	ASAP or ≤1 hour	25 °C	(2)
Wound (Abscess)				
Closed (Deep)	Aspirate preferred, Swab acceptable	ASAP or ≤1 hour	25 °C	Indicate collection site. (2) See page 25.
Open (Superficial)	Aspirate preferred, Swab acceptable	ASAP or ≤1 hour	25 °C	Indicate collection site. (2)

Note: For wound (abscess), 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. Also collect one specimen on rayon / dacron tipped, non-wooden shaft applicator in an anaerobic transport culture (A.C.T. I) see page 14 and 27. (2)



M4-RT Transport Media and 2 Dacron Swabs

Intended use:

Transport of: Chlamydia culture (routine or sexual investigations) and
 Routine Viral Cultures (most specimen types and tissue)
 (Note: This transport media **WILL NOT** support bacterial or other microbial organisms!)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Chlamydia Culture	Dacron swab or ≥1 ml aspirate or fluid in an M4-RT transport media.	ASAP or ≤ 15 minutes	4 °C	Indicate specimen source. Scrapings and tissue samples are acceptable specimens as well. Used for routine or sexual investigations. (2)
Viruses All specimen types Except Blood, BAL, CSF, Sputum, Stool, and Urine	Dacron swab or ≥1 ml aspirate or fluid in an M4-RT transport media.	ASAP or ≤ 15 minutes	4 °C	Please indicate specimen source. (2)
Tissue	≥ 0.5 g, add to an M4-RT transport media.	ASAP or ≤ 15 minutes	4 °C	Please indicate specimen source. (2)



M4 Transport Media and 2 Dacron Swabs

Intended use:

Transport of: Mycoplasma / Ureaplasma (cultures or antibody EIA) and Routine Viral Cultures (sputum, BAL or Nasopharynx aspirate or washings)

(Note: Other than Mycoplasma / Ureaplasma, Viruses and Chlamydia, this transport media **WILL NOT** support bacterial or other microbial organisms!)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Mycoplasma / Ureaplasma All sources for culture except blood	Dacron swab or ≥ 1 ml fluid in an M4 transport media	ASAP or ≤ 15 minutes	4 °C	Indicate specimen source. Usual sites of collection are respiratory, urogenital. Other sites may include body fluids and wounds/ abscesses (2,4) Test is a sendout to a Reference Laboratory. (4)
Blood, Antibodies, EIA	10 ml	ASAP or ≤ 15 minutes	4 °C	Collect blood in a 10 mL Red Top Tube (no additives). Indicate desired antibody testing for IgG, IgM or both. Test is a Sendout to a Reference Laboratory. (4)
Viruses Sputum, Bronchoalveolar Lavage or Nasopharynx Aspirate or washing	Dacron swab or ≥ 1 ml fluid in an M4 transport media	ASAP or < 15 minutes	4 °C	Please indicate specimen source. Specimen is stable at 4 °C. Test is a sendout To a Reference Laboratory. (4)

Viral Cultures from Blood



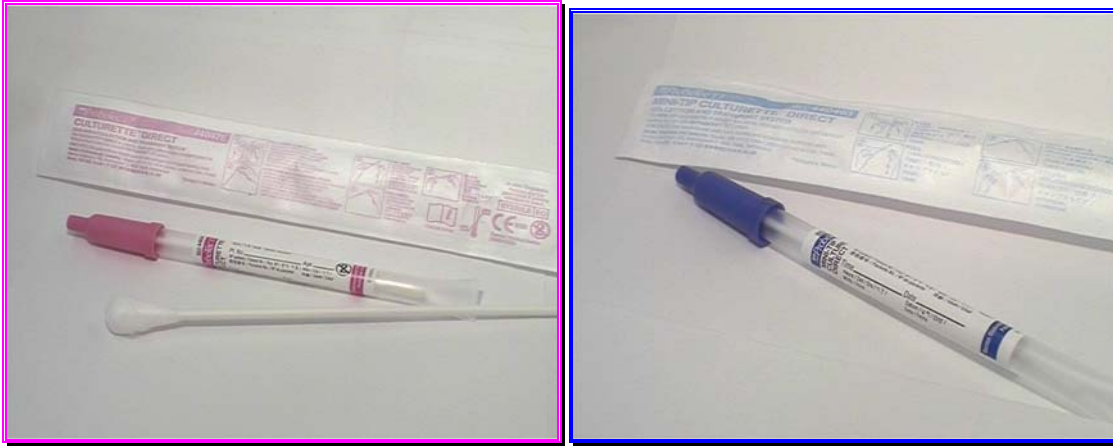
Collect Blood in 3.0 ml Sodium Heparin Green Top Tube

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
All Viruses, except CMV Blood / Bone Marrow				
Adult	3.0 ml (minimum), may need to collect 2 tubes	ASAP ≤ 1 Hour	4 °C	Do not use M4 or M4-RT viral transport media. Do not freeze. Test is a send-out to a Reference Laboratory. (4)
Pediatrics	3.0 ml (minimum), may need to collect 2 tubes	ASAP ≤ 1 Hour	4 °C	Do not use M4 or M4-RT viral transport media. Do not freeze. Test is a send-out to a Reference Laboratory. (4)



Collect Blood in 9.0 ml EDTA Lavender Top Tube

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Cytomegalovirus (CMV)	4.0 ml (minimum)	ASAP ≤ 1 Hour	4 °C	Culture not performed on blood for CMV. Testing of CMV done by hybrid capture or PCR. Test is a sendout to a Reference Laboratory. (4)



BD ProbeTec ET Culturette Direct (Female and Male)

Intended use:
Chlamydia/GC by DNA

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Chlamydia	ProbeTec swab	ASAP ≤ 15 minutes	4 °C	Indicate specimen source. (2)
GC (N. gonorrhoeae)	ProbeTec swab	ASAP ≤ 15 minutes	4 °C	Indicate specimen source. (2)



Anaerobic Transport Media (A.C.T. I)

Intended use:

Anaerobic cultures for Fluids, Tissues, Wounds,
Ear (inner), Genital (Cul-de-sac, endometrial, bartholin)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Fluids Abdominal, amniotic, ascites, bile, joint, paracentesis, pericardial, peritoneal, pleural, synovial, thoracentesis	≥ 1 ml	ASAP or ≤ 15 minutes	25 °C	Specimen should be submitted as an aspirate or fluid. (2) After sterilizing the cap, inject fluid directly through the top of the A.C.T. I media.
Genital Cul-de-sac	≥ 1 ml or swab	ASAP or ≤ 1 hour	25 °C	Aspirate or fluid submission preferred. (2) After sterilizing the cap, inject fluid directly through the top of the A.C.T. I media.
Endometrial	≥ 1 ml or swab	ASAP or ≤ 1 hour	25 °C	Aspirate or fluid submission preferred. (2) After sterilizing the cap, inject fluid directly through the top of the A.C.T. I media.
Bartholin	≥ 1 ml or swab	ASAP or ≤ 1 hour	25 °C	Aspirate or fluid submission preferred. (2) After sterilizing the cap, inject fluid directly through the top of the A.C.T. I media.

Specimen type	Device/ volume	Transport time	Transport temp. (°C)	Comments
Tissue	≥ 0.5 g, add sterile saline to sample to prevent drying.	ASAP or ≤ 15 minutes	25 °C	Always submit as much Tissue as possible. Never submit a swab that has simply been rubbed over the surface. For quantitative study, a 2 x 1 cm sample is appropriate, i.e., about 500 mg Some legionellae may be Inhibited by saline. (2)
Wound (Abscess) Closed (Deep)	≥ 1 ml or swab	ASAP or ≤ 1 hour	25 °C	Sampling of the surface area can introduce colonizing bacteria not involved in the infectious process. See page 27. (2)

Note: For wound (abscess), tissue or fluid is always superior to a swab specimen. If a swab must be used, use rayon / dacron tipped non-wooden shaft applicator and place in above anaerobic transport medium (A.C.T. D). Also, collect 2 cultrettes (modified Stuart's medium) for routine aerobic culture and Gram stain. See page 27. (2)



Blood Transport Tubes with SPS (sodium polyarethol sulfonate)

Intended use:
AFB Blood Cultures Only

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Acid-Fast Bacilli (AFB)				
Blood				
Routine AFB	≥ 1 ml	ASAP or ≤1 hour	25 °C	Indicate specimen source. Collect in SPS or heparin as an anticoagulant; EDTA or coagulated blood is not acceptable.(2)
<i>M. avium</i> complex (MAC)	≥ 1 ml	ASAP or ≤1 hour	25 °C	Indicate specimen source. Usually in immunocompromised patients, particularly those with AIDS. Collect in SPS or heparin as an anticoagulant; EDTA or coagulated blood is not acceptable.(2)
<i>M. avium</i> – <i>M. intracellulare</i> (MAI)	≥ 1 ml	ASAP or ≤1 hour	25 °C	Indicate specimen source. Usually in immunocompromised patients, particularly those with AIDS. Collect in SPS or heparin as an anticoagulant; EDTA or coagulated blood is not acceptable.(2)



Para-Pak ECOFIX and Para-Pak 10% Formalin

Please label specimen container before giving it to the patient.

Intended use:
Preservative and transport for Ova & Parasites (stool)

Specimen type	Device/volume	Transport time	Transport temp (°C)	Comments
Feces Ova & Parasites (O&P) Screen and Giardia EIA	Add specimen to line on each container. Do not fill <u>entire</u> container.	ASAP or ≤ 1 hour	25 °C	If testing for routine stool culture, Stool WBC, <i>C. difficile</i> , <i>H. pylori</i> or Rotavirus, please submit fresh, non-preserved stool, in sterile specimen container. (2) For Giardia EIA, the specimen is stable at 4 °C for 1 week. (2) Collect a total of 2 specimens, 1 specimen every other day. If patient has persistent symptoms suggesting intestinal parasitism, is immunodeficient, and/or has history of travel from tropical areas, it is recommended that specimens be collected over a 7-10 day period. This would maximize the recovery of the protozoa.

Ova & Parasite Extended Panel



Para-Pak ECOPIX and Para-Pak 10% Formalin

Please label specimen container before giving it to the patient.

Panel includes:

Routine O & P Panel Screen,
Cyclospora, Cryptosporidium,
Isospora, Microsporidia,
and Sarcocystis

Transport medium used:

Submit stool in Ecofix and
10% Formalin vials (see below)

Number submitted:

2

<u>Specimen type</u>	<u>Device/ volume</u>	<u>Transport time</u>	<u>Transport temp (°C)</u>	<u>Comments</u>
Ova & Parasite Extended Panel: Routine O & P Screen, Cyclospora, Cryptosporidium, Isospora, Microsporidia, and Sarcocystis	Add specimen to line, do not fill <u>entire</u> container	ASAP or ≤ 1 hour	25 °C	Add stool to ECOPIX and 10% Formalin vial. Collect a total of 2 specimens, 1 specimen every other day. If patient has persistent symptoms suggesting intestinal parasitism, is immunodeficient, and/or has history of travel from tropical areas, it is recommended that specimens be collected over a 7-10 day period. This would maximize the recovery of the protozoa.



Para-Pak C & S (For Culture and Sensitivity Only)

Please label specimen container before giving it to the patient.

Intended use:

Preservative and transport for Enteric Pathogens (bacteria) in stool,

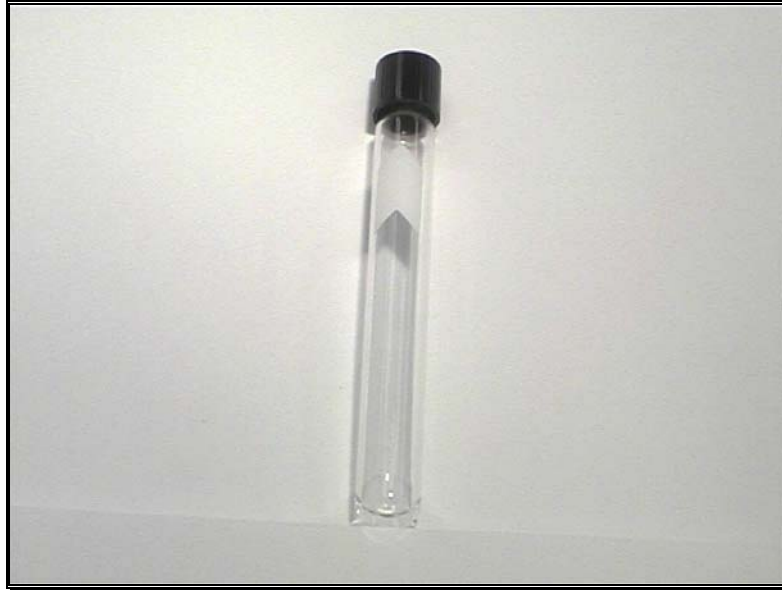
Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Feces Routine culture	Add specimen to line on container. Do not fill <u>entire</u> container.	ASAP or ≤ 1 hour	25 °C	Do not perform routine stool cultures for patients whose length of stay was >3 days and the admitting diagnosis was not gastroenteritis. Culture and toxin tests for <i>C. difficile</i> should be considered for these patients. (2) If testing for Stool WBC, <i>C. difficile</i> , <i>H. pylori</i> or Rotavirus, please submit fresh non-preserved stool, in sterile specimen container. (2)



**Vacutainer Brand Urine C&S Transport Kit -
Contains Boric Acid & Sodium Formate**

Intended use:
Preservative and Transport of Urine Cultures & Sensitivity Only

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Urine Midstream	Urine transport kit	ASAP or ≤ 48 hours	25 °C	Kit contains preservative to prevent bacterial overgrowth. (2)
Straight catheter	Urine transport kit	ASAP or ≤ 48 hours	25 °C	Kit contains preservative to prevent bacterial overgrowth. (2)
Indwelling catheter	Urine transport kit	ASAP or ≤ 48 hours	25 °C	Kit contains preservative to prevent bacterial overgrowth. (2)
Suprapubic needle Aspiration (3)	Urine transport kit	ASAP or ≤ 48 hours	25 °C	Kit contains preservative to prevent bacterial overgrowth. (2)



Sterile tube w/0.5-1.0ml of Sterile Saline with Swabbed Specimen

Intended use:
Trichomonas, Yeast, Clue Cells, Fungal Elements,
Protozoan Elements or Other Motile Organisms

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Stool	Swab and Sterile Saline	ASAP or ≤ 15 minutes	25 °C	See below. (2)
Vaginal	Swab and Sterile Saline	ASAP or ≤ 15 minutes	25 °C	See below. (2)

The saline-wet-mount preparation may be used for the detection of protozoan cyst or trophozoites and helminth eggs or larvae directly from liquid stools and from stool concentrates. Vaginal secretions may be examined by this method for the direct detection of fungi, clue cells, and *Trichomonas vaginalis*. It is important not to prepare specimens that are too thick because structures may be undetectable in the debris and background material. Conversely, mounts that contain insufficient sample suffer from poor sensitivity. Slides are examined with the light microscope for characteristic morphology and motility of organisms. (2)

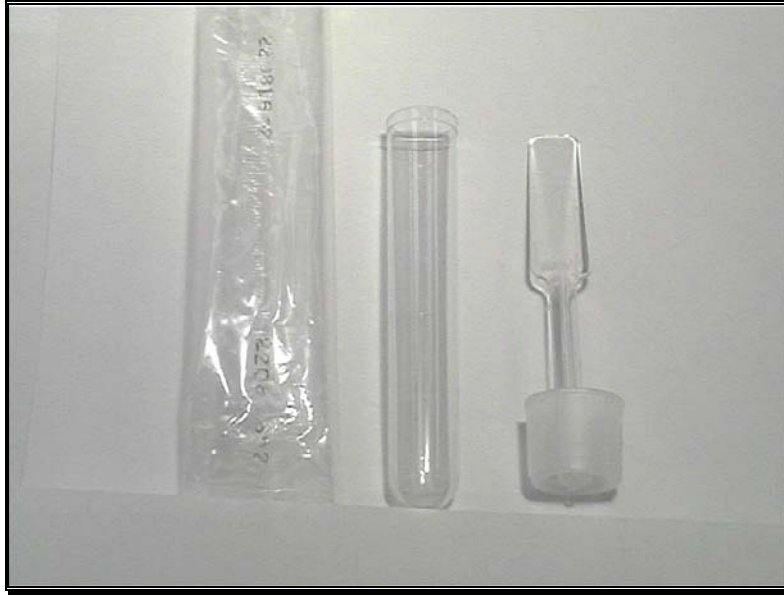


CSF Sterile Graduated Manometer Tube Kit (Tube #2)

Intended use:
 Cerebrospinal Fluid Culture, Cryptococcal Antigen (Ag) ,
 India Ink (detect *Cryptococcal neoformans* capsule)(IINK),
Legionella spp., Bacterial Antigen Detection Panel (BAD), Viral Cultures

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Bacterial Antigen Detection Panel (BAD) Panel includes detection for: <i>H. influenzae</i> , <i>N. meningitis</i> ACY, <i>N. meningitis</i> B/E, <i>S. pneumoniae</i> , Group B Strep (<i>S. agalactiae</i>)	≥ 1-2 ml	ASAP or ≤ 15 minutes	25 °C	Submit CSF (tube #2) to microbiology. (2)
CSF culture	≥ 1-2 ml Minimum amt. required: Bacteria, ≥ 1 ml Fungi, ≥ 2 ml AFB, ≥ 2 ml Virus, ≥ 1 ml	ASAP or ≤ 15 minutes	25 °C for bacteria and fungi, NEVER REFRIGERATE. Refrigerate if testing for viruses only.	Obtain blood for culture as well. If only 1 tube of CSF is collected, it should be submitted to microbiology first; otherwise submit tube # 2. (1, 2)
Cryptococcal Antigen (Ag)	≥ 0.5 ml	ASAP or ≤ 15 minutes	25 °C	Submit CSF (tube #2) or serum to microbiology. If test is positive, laboratory personnel will do a titer. (2)
India Ink (IINK)	≥ 1.0 ml	ASAP or ≤ 15 minutes	25 °C	Submit CSF (tube #2) to microbiology. (2)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
<i>Legionella</i> spp.	≥ 1 ml	ASAP or ≤1 hour	25 °C	CSF for <i>Legionella</i> is a sendout to a Reference Laboratory. (4) Test method by indirect immunofluorescence assay. (2,4)
Viruses	≥1 ml	ASAP or ≤ 15 minutes	4 °C	Frequently isolated: coxsackievirus, mumps virus, echovirus, Less frequently isolated: arbovirus, HSV, LCMV, rabies virus. (2) All CSF viral cultures are processed at Public Health Dept. (2)



Pinworm Paddle

Intended use:
Pinworm (*Enterobius vermicularis*)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Pinworm	Pinworm paddle Kit; no preservative, no minimum volume required.	≤ 24 hours	25 °C	Common parasites include: <i>Enterobius vermicularis</i> , <i>Taenia</i> spp. Specimens are best obtained at 10 to 11 p.m. or upon waking and before a bowel movement or bath. Wash hands after collection. Specimen collection should continue for ≥ 6 consecutive days before patient is considered infection-free. (2)



Specimen Trap 40cc (Luken's Trap)

Intended use:

Respiratory cultures (sputum induced, BAL, bronchial brush or wash, Tracheal aspirate),
 AFB Sputum, *Bordetella pertussis*, *Pneumocystis carinii* pneumonia (PCP), Viral culture,
 Respiratory Syncytial Virus (RSV), *Legionella* spp., Fungal Culture (sputum induced and BAL)

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Acid-Fast Bacilli (AFB) Sputum	5-10 ml	ASAP or ≤ 1 hour	25 °C	Specimen is stable at 4 °C. (2)
Respiratory, lower Sputum, Induced Bacterial	≥ 1 ml	ASAP or ≤ 1 hour	25 °C Do not refrigerate.	For pediatric patients unable to produce a specimen, a respiratory therapist should collect a specimen via suction. The best specimen should have < 10 spumous cells per 100x field. (1)
Fungal	3-5 ml	ASAP or ≤ 1 hour	25 °C	Refrigerate if specimen is not cultured within 2 hours. (3, pg. 264) (1, 2)
Bronchoalveolar Lavage, bronchial Brush or wash, Tracheal aspirate	≥ 1 ml; 3-5 ml for fungi	ASAP or ≤ 1 hour	25 °C	A total of 40 to 80 ml of fluid is needed for quantitative analysis. For quantitative analysis of brushings, place brush into 0.5 ml of Trypticase soy broth. (1, 2)

Specimen type	Device/ volume	Transport time	Transport temp. (°C)	Comments
Respiratory, lower (Cont.) Respiratory Syncytial Virus (RSV)	5-10 ml	ASAP or ≤15 minutes	4 °C	Specimen should be sent to the laboratory on ice. Nasopharyngeal aspirates or washing are a good specimen source. Other specimen collection include nasopharyngeal swabs. (2, 3)
<i>Pneumocystis carinii</i> pneumonia (PCP)	≥ 1 ml	ASAP or ≤1 hour	25 °C	Specimen collection via sputum induced, tracheal aspirate fluid, BAL, trans-bronchial biopsy (TBBx), bronchial brushing, pleural fluid, tissue from open thorax lung biopsy. (2)
<i>Legionella</i> spp.	≥ 1 ml	ASAP or ≤1 hour	25 °C	Specimen collection via sputum induced, tracheal aspirate fluid, BAL, trans-bronchial biopsy (TBBx), bronchial brushing, blood, pleural fluid, tissue from lung biopsy. (2, 4) CSF and blood may be used in detecting antibodies. Urine may be used in detecting antigens. All specimen types are sendouts to a Reference Laboratory. (4)
Viruses, Bronchoalveolar Lavage, Nasopharynx Aspirate or wash	≥1ml	ASAP or ≤ 15 minutes	4 °C	Frequently isolated: RSV, influenza virus, para-influenza virus, rhinovirus, enterovirus All specimens are sendout to a Reference Laboratory except for RSV. (2,4)
Respiratory, upper <i>Bordetella pertussis</i>	≥ 1 ml or direct inoculation of charcoal blood agar, must be kept in moist environment	ASAP or ≤15 minutes	25 °C	Specimen collection from the upper respiratory tract via nasopharyngeal swabs. higher yields are obtained by nasopharyngeal aspirates. (2)

Culture Deep Wound Panel

Collection & Transport System



Anaerobic Transport (A.C.T. I)

And CultureSwab (modified Stuart's)

And CultureSwab (modified Stuart's)

Panel includes:

Anaerobic culture

Aerobic culture

Gram stain

Transport medium used:

Submit fluid or rayon / dacron tipped non-wooden shaft applicator in anaerobic transport media, A.C.T. I (see below)

Submit CultureSwab (see below)

Submit CultureSwab (see below)

Number submitted:

1

1

1

Note: A total of three items should be submitted to the microbiology lab. If a Gram stain culturette can not be collected, the Gram stain can be obtained from the aerobic culturette. (2)

<u>Specimen type</u>	<u>Device/ volume</u>	<u>Transport time</u>	<u>Transport temp (°C)</u>	<u>Comments</u>
Wound (Abscess) Closed (Deep)	Swabs (aerobic and anaerobic)	ASAP or ≤ 1 hour	25 °C	Indicate collection site. (2)

Note: Tissue or fluid is always superior to a swab specimen. (2)

Culture Body Fluid Panel

Collection & Transport System



Anaerobic Transport (A.C.T. I)

And

Sterile specimen cup

Panel includes:

Anaerobic culture

Aerobic culture and
Gram stain

Transport medium used:

Submit fluid in anaerobic transport
media, A.C.T. I (see below)

Submit fluid (see below)

Number submitted:

1

1

Note: A total of two items should be submitted to the microbiology lab. (2)

<u>Specimen type</u>	<u>Device/ volume</u>	<u>Transport time</u>	<u>Transport temp (°C)</u>	<u>Comments</u>
Fluids Abdominal, amniotic, ascites, bile, joint, paracentesis, pericardial, peritoneal, pleural, synovial, thoracentesis	≥ 1 ml	ASAP or ≤ 15 minutes	25 °C	Pericardial fluid and fluids for fungal cultures ≤ 24 hours at 4 °C. Amniotic and culdocentesis fluids should be transported in an anaerobic system and need not be centrifuged prior to Gram staining. After sterilizing the cap, inject fluid directly through the top of the A.C.T. I media. (2)

Culture Biopsy/Tissue Panel

Collection & Transport System



Anaerobic Transport (A.C.T. I)

And

Sterile specimen cup

Panel includes:

Anaerobic culture

Aerobic culture and
Gram stain

Transport medium used:

Submit tissue in anaerobic culture
transport A.C.T. I (see below)

Submit tissue in sterile specimen
container (see below)

Number submitted:

1

1

<u>Specimen type</u>	<u>Device/ volume</u>	<u>Transport time</u>	<u>Transport temp (°C)</u>	<u>Comments</u>
Tissue	≥ 0.5 g Sterile saline may need to be added to prevent drying	ASAP or ≤ 15 minutes	25 °C	Always submit as much tissue as possible. Never submit a swab that has simply been rubbed over the surface. For quantitative study, a 2 x 1 cm i.e., about 500 mg is appropriate. Some legionellae may be inhibited by saline. (2) Submit tissue into A.C.T. I media as quickly as possible to limit exposure of obligate anaerobic bacteria to oxygen.



Dacron or Rayon or Flocked NP swab
 Charcoal Blood Agar
 Sterile Tube (for aspirate only)

Intended use:

Nasopharyngeal swabs (NP) are the specimen of choice and should be collected as soon as possible after the onset of illness, preferable prior to antibiotic therapy. Detection of pertusis will be performed by PCR and culture if requested. Nasopharyngeal aspirates may be submitted if unable to obtain NP swab.

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Pertusis Culture	Dacron swab or ≥ 1 ml fluid in Sterile Tube	ASAP or ≤ 15 minutes	4 °C	Indicate specimen source. Usual sites of collection are respiratory, include Nasopharyngeal swab , NP washing and Aspirates. For NP washing If greater than 2 hours it should be store under refrigeration. For NP wire swab, tilt Patient's head back slightly And insert the swab into one of the nostrils until it reaches the posterior nares. Leave swab in place for 15-30 seconds then withdraw and swab in Charcoal blood agar plate and or place in Reagan Lowe transport Media tubes for culture and PCR. For PCR test only place swab back in

original container.

Specimen type	Device/ volume	Transport time	Transport temp (°C)	Comments
Pertusis by PCR	Dacron swab or ≥ 1 ml fluid in Sterile Tube	ASAP or ≤ 15 minutes	4 °C	Test is a Send out to a Public Health Laboratory. (4)

References

- 1) Miller, J.M., 1999. ***A Guide to Specimen Management in Clinical Microbiology***, 2nd Edition. American Society for Microbiologists, Washington D.C.
- 2) Murray, P.R., E.J. Baron, M.A. Pfaller, F.C. Tenover, and R.H. Tenover, 1999. ***Manual of Clinical Microbiology***, 7th Edition. American Society for Microbiologists, Washington D.C.
- 3) Isenberg, H.D., 1998. ***Essential Procedures for Clinical Microbiology***. American Society for Microbiologists, Washington D.C.
- 4) Quest Diagnostic - Specimen Collection Guide. March 2001. By Quest Diagnostic
- 5) Front Cover Pictures, from Top Down and Left to Right, Courtesy of:

Tenover, F.C., Ph.D., and Hirschmann, J.V., M.D., 1990, ***Interpretation of Gram Stains and Other Common Microbiologic Slide Preparations***. The Upjohn Company, Kalamazoo, Michigan.

Figure 1 – Gram Stain slide of gram-negative coccobacilli is *H influenzae*, and the oval, gram-positive diplococci are *S pneumonia*.

Leboffe, M.J., and Pierce, B.E., 1999, ***A Photographic Atlas for the Microbiology Laboratory***, 2nd Edition. Morton Publishing Company, Englewood, Colorado.

Figure 2 – Blood Agar with unidentified throat culture isolate demonstrates β -hemolysis. The clearing around the individual colonies is due to complete lysis of red blood cells.

Figure 3 – *Cryptococcus neoformans* (Lactophenol blue stain, X1350). *Cryptococcus neoformans* is characterized by spherical, encapsulated cells that range in size from 2 to 15 μ m. The capsule is faintly visible in this preparation.

Figure 4 – *Strongyloides stercoralis* rhabditiform larva in feces (X1000, D'Antoni's iodine stain). These larvae may be distinguished from hookworm larvae (which are rarely in feces) by their short buccal cavity (B).