G. Respiratory Specimens

1. General considerations
    a) Twenty-four-hour sputum collections are not recommended for culture.
    b) If *Corynebacterium diphtheriae, Arcanobacterium haemolyticum, Bordetella pertussis, N. gonorrhoeae,* legionellae, chlamydiae, or mycoplasmas are suspected, the physician should contact the clinical microbiology laboratory prior to specimen collection because special techniques and/or media are required for the isolation of these agents.

2. Lower respiratory tract
    a) Expectorated sputum
        (1) If possible, have the patient rinse mouth and gargle with water prior to sputum collection.
        (2) Instruct the patient not to expectorate saliva or postnasal discharge into the container.
        (3) Collect specimen resulting from deep cough in sterile screw-cap cup or other suitable sterile collection assembly.

    b) Induced sputum
        (1) Using a wet toothbrush, brush the buccal mucose, tongue, and gums prior to the procedure.
        (2) Rinse the patient's mouth thoroughly with water.
        (3) Using an ultrasonic nebulizer, have the patient inhale approximately 20-30 ml of 3-10% 0.85% NaCl.
        (4) Collect the induced sputum in a sterile screw-cap cup or other suitable sterile collection assembly.
    c) Tracheostomy and endotracheal aspirations
        (1) Tracheostomy is followed by colonization within 24 hours of insertion of the tube. Results must be correlated with clinical findings such as fever or infiltrate on chest X-ray. Aspirate the specimen into a sterile sputum trap.

    d) Bronchoscopy specimens
        (1) Bronchoscopy specimens include bronchoalveolar lavage, bronchial washing, bronchial brushing, and transbronchial biopsy specimens.
        (2) Pass the bronchoscope transnasally or transorally in nonintubated patients or via the endotrachael tube in intubated patients.
        (3) Wedge the tip of the bronchoscope in a segmental (for bronchial wash) or subsegmental (for bronchoalveolar lavage) bronchus.
        (4) To obtain specimens
            (a) Bronchial wash or bonchoalveolar lavage. Bronchial wash and bronchoalveolar lavage specimens are generally obtained before brushing or biopsy specimens to avoid excess blood in the recovered fluid, because blood may alter the concentration of cellular and noncellular components.
                (i) Inject sterile nonbacteriostatic 0.85% NaCl (generally 5-20 ml aliquots) from a syringe through a biopsy channel of the ronchoscope.
                (ii) Gently suction the 0.85% NaCl into a sterile container before administering the next aliquot. (In general, 50-75% of the 0.85% NaCl instilled is recovered in the lavage effluent.)
                (iii) Keep aliquots separate during collection. Combine aliquots from the same site for microbiology cultures and smears, but aliquots from separate sites (for example, right upper lobe and right lower lobe) should be combined only after consultation with the physician of record.

            (b) Bronchial brush specimens

(i) Insert a telescoping double catheter plugged with polyethylene glycol at the distal end (to prevent contamination of the bronchial brush) through the biopsy channel of the bronchoscope.

(c) Transbronchial biopsies

(i) Obtain the biopsy sample through the biopsy channel of the bronchoscope, and transport it in a sterile container with a small amount of nonbacteriostatic sterile 0.85% NaCl.

(d) Lung aspirations

(i) Use a computed tomography scan to obtain lung aspirates by inserting a needle through the chest wall into a pulmonary infiltrate. Aspirate material from the lesion. If the lesion is large or if there are multiple lesions, collect multiple specimens from representative sites. Refer to Table 1 for instructions on properly transporting specimens collected in a syringe.

(e) Lung biopsies

(i) Obtain a 1-3 cm square piece of tissue is possible. If the lesion is large or if there are multiple lesions, collect multiple specimens from representative sites. Submit in a sterile container(s) without formalin.

(5) Upper respiratory tract infections

(a) Throat (pharyngeal specimens)

(i) Submitted primarily for the detection of group A streptococci (can also be used to detect *N. gonorrhoeae, Haemophilus influenzae* [for epiglottitis].

(ii) Do not obtain throat samples if epiglottis is inflamed, as sampling may cause serious respiratory obstruction.

(iii) Depress tongue gently with tongue depressor.

(iv) Extend sterile swab between the tonsillar pillars and behind the uvula. (avoid touching the cheeks, tongue, uvula, or lips).

(v) Sweep the swab back and forth across the posterior pharynx, tonsillar areas, and any inflamed or ulcerated areas to obtain sample.

(b) Nasal swabs

(i) Submitted primarily for the detection of staphylococcal carriers.

(ii) Insert a sterile swab into the nose until resistance is met at the level of the turbinates (approximately 1 inch into the nose).

(iii) Rotate the swab against the nasal mucosa.

(iv) Repeat the process on the other side.

(c) Nasopharyngeal swabs

(i) Submitted primarily for the detection of carriers of N. *meningitidis.*

(ii) Carefully insert a flexible-wire calcium alginate-tipped swab through the nose into the posterior nasopharynx, and rotate the swab. (Keep the swab near the septum and floor of the nose.)

(d) Nasal washings

(i) Submitted primarily for RSV viral cultures and B. pertussis.

(ii) Instruct the patient not to swallow during the procedure.

(iii) With the patient's head hyperextended (approximately 70° angle), instill approximately 3-7 ml of sterile 0.85% NaCl into each nostril.

(iv) To collect material, aspirate the fluid by inserting a rubber bulb syringe into each nostril.

(v) Place the saline wash in an equal volume of viral transport medium, or transport it in a sterile container.

(e) Sinus aspirates
  (i) Using a syringe aspiration technique, a specially trained physician or an otolaryngologist will obtain material from maxillary, frontal, or other sinuses.
  (ii) Place the contents of the syringe into an anaerobic transport system, or send the specimen in the syringe.

(f) Typanocentesis fluid
  (i) Submitted primarily to diagnose middle ear infections only if previous therapy has failed.
  (ii) Clean the external canal with mild detergent.
  (iii) Using a syringe aspiration technique, the physician will obtain the fluid from the eardrum. Send the specimen in a sterile container, or send it in the syringe.
  (iv) If the eardrum is ruptured, collect exudate by inserting a sterile swab through an auditory speculum.

(g) Oral cultures
  (i) Used to prepare smears for the detection of yeast or fusospirochetal disease.
  (ii) Rinse mouth with sterile saline.
  (iii) Wipe the lesion with dry sterile gauze.
  (iv) Swab or scrape areas of exudation or ulceration.

(6) Respiratory specimen collection considerations are summarized in Table 8.

Table 8: Collection Considerations for Respiratory Specimens

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