Cytology Specimen Collection and Handling Guidelines

Proper specimen collections require that specimen integrity is maintained by proper preservative (where required), that sample identification and patient identification be clearly labeled on the specimen container or slide and that the sample is properly transported to the clinical laboratory in a timely manner. The information listed below is to assist with that objective.

**SPECIMEN COLLECTIONS - GENERAL INFORMATION**

All cytology specimens received by the laboratory must be accompanied by a completed cytology requisition that includes the following information:

- Patient’s full, legal name
- Physician(s) name
- Patient’s date of birth
- Patient’s Social Security number
- Date of specimen collection
- Source of specimen (anatomic site)
- Brief clinical history (CLIA requirement)

Cytology specimens received in the laboratory after 2:30 p.m. on weekdays and on weekends or holidays will be evaluated by laboratory staff to see that they are properly preserved to prevent cellular degeneration. Testing will be performed on the next routine processing day.

Specimen containers and slides must be labeled with the patient’s complete name and specimen source.

Information concerning specimen rejection criteria is listed in the “Specimen Collection” section of this manual.

**BODY FLUID (PLEURAL, PERITONEAL, PERICARDIAL, SYNOVIAL)**

1. Collect 10-200 ml specimen in a leak-proof, screw-top, wide-mouth container. The optimum sample size is 100 ml or larger. A cell block will be prepared if adequate material is obtained.
2. Immediately label the specimen container with the patient’s complete name and source of specimen.
3. Transport the specimen and requisition to the lab as soon as possible. If transport is delayed by more than 10 minutes, add CytoLyt™ to the specimen (ratio of 50 percent specimen and 50 percent CytoLyt).
4. If multiple tests are ordered on the same specimen (i.e., hematology, chemistry, microbiology, etc.), divide the sample into appropriate tubes before adding preservative. See the alphabetical test listing for specific sample requirements for other tests.

**BREAST-NEEDLE ASPIRATION**

1. Palpate, localize and wipe aspiration site with antiseptic.
2. Insert needle and retract piston to create suction in the needle.
3. Under constant suction, move needle tip backward and forward. If possible, direct it toward different areas.
4. Release suction before removing the needle from the lesion.
5. Rinse syringe and needle into CytoLyt solution.
6. Label the CytoLyt tube with the patient’s complete name and specimen identification.
7. Transport sample and requisition to the lab as soon as possible.

**SOLID MASS ASPIRATE AND CYST ASPIRATE**

1. Follow steps 1-5 as outlined in “Needle Aspiration.”
2. Prepare one direct smear or touch prep microscope slide of aspirate. Immediately fix in 95 percent ethyl alcohol or flood slide with spray fixative.
3. Express remainder of aspirate into CytoLyt solution. A cell block will be prepared if adequate material is obtained.
4. Immediately label the slide and specimen container with patient’s complete name and specimen identification.
5. Transport the sample and requisition to the lab as soon as possible.

**NIPPLE SECRETION**

1. Directly smear nipple discharge onto labeled slide (include source as “right nipple” or “left nipple”).
2. Immediately fix in 95 percent ethyl alcohol or flood slide with spray fixative.
3. Transport the slide and requisition to the lab as soon as possible.

**BRONCHIAL BRUSHING**

2. Label specimen container with patient’s complete name and specimen identification.
3. Bring specimens immediately to the laboratory or refrigerate.

**BRONCHIAL WASHING**

1. Collect specimen in a leak-proof, screw-top, wide-mouth container. Instill 20-60 ml of normal saline then re-aspirate the material. A cell block will be prepared if adequate material is obtained.
2. Label slide and specimen container with the patient’s complete name and specimen identification.
CSF (CEREBROSPINAL FLUID)
1 Collection of CSF is a surgical procedure. The physician should wear gloves, mask and gown for specimen collection.
2 Prepare the skin in the same manner as for blood culture.
3 Overlay the area surrounding the puncture site with sterile drapes.
4 Insert the needle. Collect the fluid into three leakproof, sterile containers. Collect an adequate amount of fluid. Minimum sample is 1 ml. The sample should be free of blood or solid material.
5 Cap the tubes tightly, label properly with the patient’s complete name and specimen identification. Submit to the laboratory immediately. DO NOT REFRIGERATE.

ESOPHAGEAL BRUSHING
1 Submit brush tip in CytoLyt.
2 Label specimen container with patient’s complete name and source of specimen.
3 Bring specimens to the laboratory immediately.

FINE NEEDLE ASPIRATION (FNA)
Fine needle aspiration procedures performed in surgery/CT/PTC should be scheduled with the cytology department between 8:00 a.m. and 4:00 p.m. so that assistance with slide preparation/fixation may be provided. Contact the pathology department at 630.933.2636 for aspirations performed after 4:00 p.m. and on weekends.

Semi-Solid Aspirate:
1 Rinse needle with CytoLyt solution.
2 Prepare air-dried slides if aspiration site is a lymph node.
3 Label specimen container and slides with the patient’s complete name and source of specimen.

Cystic Aspirate:
1 Transfer contents of syringe into CytoLyt solution.
2 Label specimen container with the patient’s complete name and source of specimen.
If cultures are required, dispense a portion of cystic fluid into a sterile container without CytoLyt solution.
If a definitive diagnosis is not achieved, a re-aspiration may provide the needed diagnostic material.

GASTRIC WASHING/BRUSHING
The patient should fast for 12 hours prior to the lavage procedure, but should drink water on the morning of the test.

Washing
Approximately 300 ml normal saline should be injected, withdrawn and re-injected several times through a #16 French gauge plastic Levin tube at about the 55 cm mark.
1 Minimum sample size is 50 ml. A cell block will be prepared if adequate material is obtained.
2 Add an equal amount of CytoLyt to the specimen. Refrigerate if unavailable.
3 Label specimen container with the patient’s complete name and source of specimen.

Brushing
1 Submit brush in CytoLyt.
2 Label specimen container with the patient’s complete name and source of specimen.

PAP SMEAR - ENDOCERVICAL SAMPLE
Specimen should be collected (preferably) two weeks after the first day of the last menstrual period and definitely not during menstruation.
Patient should be instructed to not use vaginal medication, contraceptives, lubricants or douches 48 hours prior to the test and to refrain from intercourse the night before the examination.
Specimen must be accompanied by a cytology requisition with the following information:
• Patient’s name and address
• Social Security number (must be included to comply with state regulations)
• Date of birth
• Physician’s name
• Date of collection
• Source of specimen
• Clinical information: LMP (required), hormone use, hysterectomy, pregnant, previous abnormal Pap with date and cyto number, risk factors or previous cancers
• Billing information where applicable
For supplies such as PreservCyt Vial Solution, collecting devices (cytobroom brushes, plastic spatula, soft-bristled brushes) please fax an order to the supply department at 630.933.5056.

THINPREP PAP PROCEDURE:
1 Pre-label a vial of PreservCyt Solution with the patient’s complete name and collection date.
2 Obtain the gynecologic specimen from the patient in the usual manner.
3 Rinse the collection device into the vial of PreservCyt solution.
4 Send the specimen vial and cytology requisition to the laboratory for slide preparation and screening.
5 Request HPV/DNA. If results are ASCUS, HPV testing will be automatically performed.
6 Request chlamydia/GC from vial, if indicated.
SINGLE-SLIDE PAP SMEAR PROCEDURE:

1. Pre-label microscope slide with the patient’s name using a lead pencil.
2. With the endocervical brush, gently insert into the endocervix, turn 360 degrees and withdraw.
3. Prepare endocervical specimen by rolling brush, with moderate pressure over the ectocervical sample; smear both across the slide.
4. Immediately fix with Pap-Pak kit fixative, cytology spray fixative or immerse in 95 percent ethyl alcohol. Immediate fixation is necessary for optimal cellular interpretation. Spray fixatives should be held approximately 12 inches from the slide to prevent cellular artifacts.
5. Reconfirm that the slide is labeled with the patient’s name. Place slide in plastic slide mailer container after it is completely dry.
6. Request HPV/DNA. If results are ASCUS, HPV testing will be performed. A Digene kit sample is needed to test HPV/DNA when a conventional slide is submitted.

PNEUMOCYSTIS CARINII

Bronchoalveolar lavage (BAL) is the preferred specimen for Pneumocystis carinii evaluation. Other specimens include spontaneous and induced sputum, bronchial washing, bronchial brushings, transbronchial and trans-thoracic needle aspirations.

A minimum of 20 ml specimen for bronchial washing and bronchoalveolar lavage is recommended.

Submit in tightly capped, leak-proof container labeled with patient’s complete name and specimen source.

SPUTUM

Assure patient cooperation to collect an adequate and acceptable specimen. The sputum should be produced from a deep cough from the respiratory tract. The optimal time of day for sputum collection is early morning.

Induced sputum should be obtained if the patient cannot spontaneously produce a specimen. Collection of oral contents will be rejected by the laboratory.

Only one sputum should be collected and submitted per patient per day (the only exception is a post-bronchoscopy sputum specimen). Do not pool multiple samples.

1. Obtain a first morning specimen before the patient brushes his or her teeth, uses mouthwash and before eating. Instruct the patient to rinse his or her mouth with water before collecting the sample.

2. Instruct the patient to expectorate into a wide-mouth, screw-capped container. Do not ask the patient to “spit” into the container as this generally results in a saliva specimen instead of a sputum specimen.

3. If a patient is unable to produce sputum, specimen induction may be necessary. Postural drainage, saline nebulization or chest percussion may be used.

4. Label the specimen container with the patient’s complete name and specimen source.

5. Add CytoLyt immediately in equal proportion to the specimen. If CytoLyt is unavailable, refrigerate and transport immediately to the laboratory.

URINE - CYTOLOGY

An acceptable specimen may be a bladder irrigation sample or a “clean catch” voided sample, 24-hour samples will not be accepted.

If cultures are required, aliquot a portion of specimen into a sterile container or urine vacutainer tube before adding CytoLyt fixative to the cytology sample.

Bladder Urine

1. Use 50 to 75 ml of normal saline to irrigate the bladder, agitate and withdraw the sample.

2. Use a minimal amount of lubricant to prevent obscuring of cellular detail.

3. Submit in a tightly capped, leak-proof container labeled with the patient’s complete name and specimen source.

Clean-Catch Voided Urine

1. Collect a second early morning void sample for optimal results. See “Urine Collections” in the “Specimen Collection” section for detailed collection instructions. Minimum sample size is 50 ml.

2. Add an equal amount of CytoLyt to the sample immediately. Refrigerate specimen if CytoLyt is unavailable.

3. Submit in a tightly capped, leak-proof container labeled with the patient’s complete name and specimen source.

TZANCK TEST

Rupture vesicle and scrape the base of lesion to rule out viral infection.

1. Label slides using a pencil with patient’s name.

2. Spread material thinly on slides.

3. Immediately immerse slide(s) in 95 percent ethyl alcohol, spray fixative or air dry.

4. Indicate site of lesion on cytology requisition and method of fixation.