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# HISTOLOGY AND CYTOLOGY

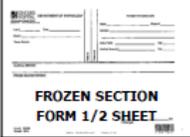
## Specimen Labeling and Requisitions Requirements

Specimens will not be processed unless:

1. All specimens labeled with: Patient Name, DOB or MR#, Date and Time of collection, Collector's initials and Specimen Source
2. All requisitions adequately completed with pertinent information regarding clinical history
3. Specimen label and requisition information match.
4. Requisition is signed by the ordering physician

### WORK INSTRUCTIONS:

Specimen Submission Requirements

Study	Type	Container	Label	Form(s)	Delivery
<b><u>FROZEN SECTIONS</u></b>	Fresh	 FROZEN SECTION FORM 1/2 SHEET	 Name, DOB, MRN, Specimen Type, Initials	Cerner: Pathology Frozen Section Request. If not on Cerner Pathology Requisition Frozen Section Form	Back Door of Pathology (Histology)
<b><u>KIDNEY BIOPSY, FLOW CYTOMETRY or SPECIAL STUDIES (i.e. lymph node; products of conception</u></b>	Unfixed (Dry)	Sterile Dampened Saline Gauze	 Name, DOB, MRN, Specimen Type, Initials	Cerner: Pathology Tissue Request. If not on Cerner: Pathology Surgical Requisition	Back Door of Pathology (Histology)
<b><u>PERMANENT (NO SPECIAL TESTING) SURGICAL TISSUE</u></b>	Fixed in Formalin	Container Larger than Specimen 10% Buffered Formalin filled to COMPLETELY COVER Tissue Ratio (20:1)	 Name, DOB, MRN, Specimen Type, Initials	Cerner: Pathology Tissue Request. If not on Cerner: Pathology Surgical Requisition	<b>Red Bin</b> for delivery to Pathology or Laboratory
<b><u>STONE ANALYSIS SPECIMEN</u></b>	Unfixed (Dry)		 Name, DOB, MRN, Specimen Type, Initials	Calculi Stone Analysis-ARUP	Tube System or Deliver to Laboratory Window - NOT HISTOLOGY

# **HISTOLOGY SPECIMEN COLLECTION AND ORDERING**

**Tissue specimens and biopsies** (e.g. placentas, renal, liver, pleural biopsies, etc.)

- Collect in clean container with at least **10x the amount of 10% Buffered Formalin** per specimen size. → **Specimens must be completely covered and floating, for adequate fixation.**
- Complete label on specimen and send to Histology lab with the following form:  
**Patient Care Units** accompanied by a Pathology requisition.  
**Operating Room** accompanied by the Operating Room Data Sheet.

**Lymph Node Studies and Frozen Sections \*\*\*\*SENT FRESH\*\*\*\***

**Do Not Add Formalin to these Specimens**

- Notify Histology at ext. 4660 before the specimen arrives.
- Label the specimen **“FRESH”** and send directly to the Histology Lab accompanied by the **O.R. Data Sheet and a Frozen Section form** → Provide O.R. room extension number to contact the operating surgeon
- Notify Histology upon arrival in the lab.

## **KIDNEY BIOPSY**

**ALL** Kidney biopsies **MUST** be received **“FRESH”** on saline dampened gauze, in a specimen container. The core sample should be a minimum of 1mm in size with 3 mm being ideal for diagnostic interpretation. The Specimen **MUST** be received with a Pathology requisition **AND** a completed Brigham and Women’s Hospital Pathology Requisition signed by the ordering doctor.

## **LIVER BIOPSY**

**ALL** Liver biopsies sent for special studies (i.e. Quantitative Iron Study; Copper study) **MUST** call Pathology prior to collection of specimen. Collect a minimum of two cores. The specimen can be either fresh or in formalin. The Liver Biopsy should measure at least a 1 cm in length, for optimal testing results. The ordering physician must enter into Cerner a Pathology Tissue request and add “Quantitative Iron needed” in the clinical information.

## **Bone Marrow Aspirations**

- Call Hematology at ext. 4650 to notify of appointment
- Complete CH Bone Marrow Biopsy and Aspirate Order.

## **PRODUCTS OF CONCEPTION**

- If **Genetic Studies** are to be performed on the tissue, the specimen **MUST** be sent to Pathology  
In the **FRESH** state **OR** in **SALINE** in an appropriate labeled plastic surgical container. (Chromosomal analysis for example)
- If **NO GENETIC STUDIES** are to be performed the specimen may be sent to Pathology in **10% formalin** in an appropriate labeled plastic surgical container.
- All products of conception sent to the Pathology department must be labeled with patient name, date of birth, date and time of collection.
- Completed pathology requisitions must accompany specimen.

## **FETUS/FETAL TISSUE**

### **If the Fetus/Baby is less than 20 weeks old:**

- The tissue is sent to the Pathology Dept. in **saline** in an appropriate plastic surgical container. **NOTE: Saline will allow for Genetic Studies if needed.**
- Label container with patient name, date of birth, date and time of collection
- Completed Pathology requisitions must accompany specimen- Order as a surgical specimen.

### **If the Fetus/Baby is over 20 weeks old:**

- Over 20 weeks the Fetus/Baby should be wrapped in a blanket and sent to the Morgue for either autopsy or disposition.
- **PLEASE CALL PATHOLOGY TO LET THEM KNOW A BABY IS BEING SENT.**

**NOTE: Any Fetus/Baby born alive or shows signs of life (regardless of how briefly) **MUST** be buried or cremated by a funeral director. Internal disposition is an option only for Fetus/Baby death under twenty week's gestation.**

# CYTOLOGY SPECIMEN COLLECTION AND ORDERING

## GENERAL

For proper interpretation, cells for cytologic examination must be properly fixed. To ensure the latter, directions for fixation must be followed explicitly.

- Fixation solutions are kept in the Histopathology Laboratory.
- Use only the fixatives recommended in the directions.
- Questions that are not answered in the following directions should be referred to the Cytotechnologist or Pathology Supervisor prior to the collection of the specimen

(Cytology- Ext: 4665)

## WORK INSTRUCTION:

Specimen Submission Requirements

Study	Type	Container		Label	Volume	Additive	Form(s)	Delivery
<b><u>BRONCHIAL WASHING</u></b>	Fresh	Luki Suction Container		Name, DOB, MRN, Specimen Type, Initials	N/A	N/A	Cerner: Pathology Non-GYN Request. If not on Cerner: Pathology Surgical Requisition Pathology Requisition	<b>Red Bin</b> for Delivery to Pathology/ Cytology or Laboratory Window
<b><u>PLEURAL, PERITONEAL, or PERICARDIAL FLUID</u></b>	Unfixed	Vacutainer Bottle or Bag; Sterile Syringe or Sterile Container		Name, DOB, MRN, Specimen Type, Initials	250-500 mL aliquot of the uniformly mixed specimen	4000 units (2000/2ml) units of heparin INJECT into vac bottle Rotate vac bottle to mix	Cerner: Pathology Tissue Request. If not on Cerner: Pathology Surgical Requisition	<b>Red Bin</b> for Delivery to Pathology/ Cytology or Laboratory Window
<b><u>PELVIC WASHING</u></b>	Unfixed	Sterile Urine Container		Name, DOB, MRN, Specimen Type, Initials	N/A	N/A	Cerner: Pathology Tissue Request. If not on Cerner: Pathology Surgical Requisition	<b>Red Bin</b> for Delivery to Pathology/ Cytology or Laboratory Window
<b><u>URINE SPECIMEN FOR CYTOLOGY</u></b>	Unfixed	Sterile Urine Container		Name, DOB, MRN, Specimen Type, Initials	3 to5 mL Required for Optimal Diagnostic Value	N/A	Cerner: Pathology Tissue Request. If not on Cerner: Pathology Surgical Requisition	<b>Red Bin</b> for Delivery to Pathology/ Cytology or Laboratory Window
<b><u>BRUSHINGS: BRONCHIAL, BILIARY OR PANCREATIC DUCT</u></b>		Brush Specimen-Cytolyt Specimen Tube Slides- 95% Alcohol Biliary or pancreatic duct brushing "FLUID" goes in Cytolyt solution <u>with</u> the brush. "Solid TISSUE" is placed into formalin.		Name, DOB, MRN, Specimen Type, Initials	N/A	N/A	Cerner: Pathology Tissue Request. If not on Cerner: Pathology Surgical Requisition	<b>Red Bin</b> for Delivery to Pathology/ Cytology or Laboratory Window

### **BLADDER IRRIGATION (WASHINGS)**

Obtain washings of the bladder with hormonal saline or Ringer's solution at the time of cystoscopy. Label specimen and send to the Cytology Laboratory immediately, accompanied by a Pathology Test Requisition Form.

### **BREAST SMEARS and FINE NEEDLE ASPIRATION FOR CYTOLOGY**

Label two or more glass slides with patient's name and other information. After aspiration has been completed, place a drop or two of the material on the centers of as many glass slides as possible, immediately covering each slide with a second clean glass slide. After all slides have been covered, gently pull the slides apart, smearing the fluid along the length of each slide. Fix slides IMMEDIATELY in 95% ethanol. The slightest air drying of the smear prior to fixation will lead to some distortion of the cell sample; therefore the smears that are fixed immediately (wet fixed) will yield the best preservation of the nuclear detail which is mandatory for accurate interpretation.

If there is any remaining material, draw saline solution into the barrel of syringe, using the same needle. Expel entire contents into specimen container of CytoLyt Fixative (about 50cc) and send it ASAP to the Cytology Laboratory for preparation, accompanied by a Cerner ordered: Pathology Non-GYN request.

Reference: Koss, LG, Diagnostic Cytology and Its Histopathologic Bases, 3rd Ed., Vol. 2 p1005; 1979.

### **CEREBROSPINAL FLUID**

The majority of specimens from the central nervous system fluid compartment are drawn via lumbar puncture. Cisternal and ventricular taps, as well as direct aspirations of cystic or solid masses also provide very useful cellular samples. Knowledge of the collection method and site is essential to the interpretation of the cellular collection. Cellular deterioration is the main problem encountered when delay occurs. Adequacy of specimen volume is of equal significance to the diagnostic value; 3-5cc is preferred. Use second or third sample drawn for cytology. Label and send to the Cytology Laboratory immediately, accompanied by a Cerner ordered: Pathology Non-GYN request.

## **GASTRO-INTESTINAL SPECIMENS**

### **COLONIC BRUSHINGS**

Brushings of mucosal abnormalities at the time of fiber optic examination of the colon are the generally accepted procedure in patients suspected of harboring a colonic lesion. Specimen is smeared on clean glass slides and fixed **immediately** by placing in 95% ethyl alcohol. Label and send to the Cytology Laboratory accompanied by a Cerner ordered: Pathology Non-GYN request.

### **COLONIC WASHINGS**

Preparation of the patient is important since the colon must be free of fecal material. The patient is instructed to take 2 oz. of castor oil 12 hours prior to the collection period and to have only a very light meal on the morning of collection. Warm soap suds enemas are given until the returns are clear. Two to three hours later the specimen is collected by colonic irrigation. Five hundred (500) ml. of warm (37° C.) saline or Ringers solution is instilled with the patient on the left decubitus position. Three to five (3-5) minutes later the fluid is collected. Label and send specimen to the Cytology Laboratory accompanied by a Cerner ordered: Pathology Non-GYN request.

### **DUODENAL and GASTRIC WASHINGS**

The patient should not have eaten food in the previous eight hours. The gastric tube should be passed to the 70 cm mark without the use of any lubricants except glycerin. Small sips of saline (not water) may be used to aid in passing the tube. Two hundred fifty (250) ml of Ringers solution or physiologic saline are then introduced into the stomach in small proportions, aspirated and discarded. If the patient has pyloric obstruction, several such lavages may be necessary. Introduce 250 ml of Ringers solution or saline and re-aspirate and reintroduce the fluid several times using a 50 or 100 ml. syringe and with the patient in different positions (prone, supine, left side, right side). The entire gastric contents are then aspirated. Send specimen to the Cytology Laboratory without delay, accompanied by a Cerner ordered: Pathology Non-GYN request.

### **ESOPHAGEAL WASHINGS**

Material is best obtained by direct esophagoscopy. Small amounts of physiologic saline are injected through the esophagoscope and aspirated. Place the aspirate immediately in CytoLyt fixative. Label and send to the Cytology Laboratory immediately, accompanied by a Cerner ordered: Pathology Non-GYN request.

### **RECTAL WASHINGS**

Preparation is the same as for the Colonic Washings. Two to three hours after the cleansing enema, 10 ml of normal saline is introduced through the sigmoidoscope and after 30 seconds is aspirated by strong suction. Label and send to the Cytology Laboratory accompanied by a Cerner ordered: Pathology Non-GYN request.

### **GYNECOLOGICAL SMEARS**

- Patient should not be douched for 24 hours before genital smears are obtained.
- Preferably, smears should not be taken during menstrual bleeding.
- No lubricant should be used.
- Requests for special testing (HPV; GC Chlamydia; Trichomonas) must be requested at time of Pap testing.

**NOTE: HPV, GC Chlamydia and Trichomonas are now performed in Concord Hospital Laboratory directly off of the Thin Prep Pap Vial, on the Hologic Panther System.**

### **THIN PREP LIQUID BASED PAP PREPARATION COLLECTION TECHNIQUE**

<http://www.thinprep.com/pdfs/man-01704-001.pdf>

#### **Sample Collection Technique using Broom-Like Collection Device:**

1. Obtain a sample from the cervix using a broom-like device. Insert the Broom-like collection device into the endocervical canal, push gently and rotate in a clockwise direction five (5) times
2. Rinse the broom collection device immediately into the Thin Prep Vial by pushing the broom into the bottom of the vial ten (10) times forcing the bristles apart. As a final step, swirl the broom vigorously to further release material into the vial. **Discard the collection device.**
3. Cap the vial tightly.
4. Label with patient's name and date of birth. Send specimen to the Laboratory for processing, accompanied by a Cerner ordered: Pathology GYN request.

#### **Sample Collection Technique using Brush/Spatula Collection Device:**

1. Obtain a sample from the cervix using the brush/spatula collection device. Insert the contoured end of the plastic spatula and rotate 360 degrees around the entire exocervix while maintaining tight contact with the exocervical surface.
2. Rinse the spatula immediately into the Thin Prep vial by swirling vigorously in the vial ten (10) times. **Discard the spatula.**
3. Obtain a sample from the endocervix using the endocervical brush device. Insert the Brush device into the cervix until only the bottom most fibers are exposed. Slowly Rotate ¼ to ½ turn in ONE direction. **DO NOT OVER ROTATE.**
4. Cap the vial tightly
5. Label with patient's name and date of birth. Send specimen to the Laboratory for processing, accompanied by a Cerner ordered: Pathology GYN request.
- 6.

### **PAP URINE**

NOTE: Obtain separate specimens for routine and bacteriological examination. Voided urine is preferable; however, catheterized urine is acceptable. Three morning non-initial samples of urine, each of about 50 to 100 ml. obtained on consecutive days are recommended. Hydration of patients by forced intake of fluids (1 glass of water every 30 minutes for a 3 hour period) is advocated. Label specimen and send to the Cytology Laboratory immediately, accompanied by a Cerner ordered: Pathology GYN request.

### **EUS PROCEDURE**

Endoscopic ultrasound (EUS) is a procedure that uses sound waves to create visual images of the internal parts of the body. The Endoscopic ultrasound (EUS) allows the surgeon to perform a biopsy to diagnose a tumor, plan treatment, and to check for recurrence after treatment. This is done using fine needle aspiration (FNA). During this process, a thin needle is placed through the endoscope and directed into the mass or surrounding lymph nodes to obtain a biopsy specimen.

The needle is withdrawn from the area being biopsied and the aspirated matter is expressed onto glass slides, the number of slides depending on the amount of aspirate. Fix half of the total smears collected in 95% alcohol. Air dry the remaining number of smears. The needle and syringe may be washed with sterile normal saline and expelled into a specimen container containing CytoLyt fixative on the first pass. All Subsequent passes are washed in formalin and the contents sent for examination. Label and send specimen immediately to the Cytology Laboratory accompanied by a Cerner ordered: Pathology GYN request. **NOTE:** If a tissue core is taken at time of biopsy the tissue core should be placed into 10% Formalin and sent to Pathology accompanied by a Cerner ordered: Pathology Tissue request.

### **LUNG ASPIRATE and NEEDLE BIOPSY**

Histologic and Cytologic study of pulmonary tissue are often made on material collected with the aid of a wide-bore needle or a trocar and cannula. Needle biopsy of the lung is most commonly performed to investigate lesions that are inaccessible to the bronchoscope and do not desquamate into the bronchial tree. Image-intensifier television fluoroscopy is used to monitor pulmonary aspiration. The patient is positioned horizontally on an adjustable table. The point on the skin from which the underlying lesion can be penetrated is marked. The skin, chest wall and pleura are anesthetized. With television fluoroscope guidance, the needle is inserted close to the upper margin of a rib (to avoid the intercostal artery) and introduced into the lesion.

Care must be taken to avoid large blood vessels and bronchi. The patient is instructed to breathe normally. When the tip of the needle is in the desired position, the operator rotates it so as to loosen small tissue fragments. A 10-20 cc. syringe is attached to the needle, and the loosened tissue is aspirated while the patient holds his/her breath. The needle is withdrawn from the chest, and the aspirated matter is expressed onto glass slides, the number of slides depending on the amount of aspirate. Fix half of the total

smears collected in 95% alcohol. Air dry the remaining number of smears. The needle and syringe may be washed with sterile normal saline and expelled into a specimen container containing CytoLyt Fixative and the contents sent for examination. Label and send specimen immediately to the Cytology Laboratory accompanied by a Cerner ordered: Pathology NON-GYN request for aspirates and a Pathology Tissue Request for needle biopsy.

### **EBUS PROCEDURE**

The Endobronchial ultrasound (EBUS) is used to diagnose and stage lung cancer, and to determine if the disease has spread to other parts of the body, such as the lymph nodes. This technique allows the surgeon to obtain real-time images in and around the lungs and to identify difficult-to-reach tumors. The Endobronchial ultrasound (EBUS) is also used to obtain a biopsy of tissue or fluid sample from the lungs and surrounding lymph nodes of the chest.

During an EBUS procedure, a thin, flexible instrument called a bronchoscope is fitted with an ultrasound device and guided through the patient's mouth and trachea, a thin needle is placed through the bronchoscope and directed into the mass or surrounding lymph nodes to obtain a biopsy specimen.

The needle is withdrawn from the area being biopsied and the aspirated matter is expressed onto glass slides, the number of slides depending on the amount of aspirate. Fix half of the total smears collected in 95% alcohol. Air dry the remaining number of smears. The needle and syringe may be washed with sterile normal saline and expelled into a specimen container containing CytoLyt fixative on the first pass, all other passes are expelled into formalin and the contents sent for examination. Label and send specimen immediately to the Cytology Laboratory accompanied by a Cerner ordered: Pathology Non-GYN Request. **NOTE:** If a tissue core is taken at time of biopsy the tissue core should be placed into 10% Formalin and sent to Pathology accompanied by a Cerner ordered: Pathology Tissue Request.

### **PULMONARY SPECIMENS**

#### **BRONCHIAL ASPIRATES OR SALINE WASHINGS**

Bronchial aspirates are obtained by suction during bronchoscopic procedures.

***Bronchial Washings:*** With the bronchoscope in position, the patient is placed on the table in such manner that the suspicious lung is dependent. The tip of the bronchoscope is placed as close as possible to the area to be investigated. About 10 ml. of normal saline is instilled in small portions of 2-3 ml. at a time and re-aspirated while the patient is made to cough. The flexible tip of the aspirator may be placed also in the opening of some of the smaller bronchi and the procedure repeated. All the cellular material is collected in a clef collection. The collection tube should be rinsed thoroughly with saline and the rinsing's added to the specimen. The specimen must be labeled and send to the Cytology Laboratory immediately, accompanied by a Cerner ordered: Pathology Non-GYN request.

## **PLEURAL, PERITONEAL and PERICARDIAL FLUIDS**

Fluids are drawn by needle aspiration from pleural, pericardial and peritoneal spaces. Specimens may also be obtained by needle insertion and irrigation of cavities or by simple suction of free fluid encountered at surgical exploration of cavities. Do not add fixative to the unprocessed specimen. An anticoagulant should be added to prevent fluid clotting - 5000 units of heparin per liter of fluid is recommended. Rotate container to properly mix specimen and anticoagulant. A 250 - 500 ml. aliquot of the uniformly mixed specimen is sufficient for evaluation. Label the container. Complete a Cerner ordered: Pathology Non-GYN request. It is essential to include the site, method of collection and clinical history.

- Send specimen to the Cytology laboratory without delay.
  - Refrigerate specimen if delay is unavoidable.

## **SPUTUM**

Instruct the patient to expectorate directly into a clean container. Morning specimens resulting from overnight accumulation of secretions yield the best diagnostic results. Three specimens on three successive days should be collected to ensure a maximum of diagnostic accuracy. The patient must be carefully instructed not to spit into the container without a deep cough, since saliva is of no diagnostic value. Specimen must be labeled and sent to the Cytology Laboratory immediately, accompanied by a Cerner ordered: Pathology Non-GYN request.

## **PCP STAIN**

PCP Stain is a Special Stain used to rule out Pneumocystis Carinii in Bronchial Specimens and Sputum Specimens. The Bronchial Specimen or the Sputum Specimen should be sent to the Cytology Dept. in the Fresh State. The Physician should write the orders for PCP Stain to be done at the time of ordering which should accompany the specimen to the lab.

## **RENAL ASPIRATION BIOPSIES**

Renal aspiration biopsies are used almost exclusively in the diagnosis of lesions detected by radiologic methods. Most are performed with the aid of television fluoroscopy or ultrasound.

When the target has been exactly located by fluoroscopy or ultrasound, the patient is positioned prone for television fluoroscopy with one or two pillows under the abdomen. Approximately ten minutes before the biopsy is begun, intravenous pyelography is performed. When the renal pelvis has been visualized, the site for aspiration biopsy is selected. The skin is cleansed and the underlying tissues are anesthetized. A fairly thick needle (external diameter 1.2mm. length 10cm.) containing a mandrin is inserted 5 to 8 cm. A series of aspirations takes place and an abundance of blood or fluid is obtained

and deposited on clean glass slides. Slides are fixed immediately by placing in coplin jar containing 95% ethyl alcohol. Rinse all aspirates into specimen container containing CytoLyt fixative. Label and send specimen immediately to the Cytology Laboratory accompanied by Cerner ordered: Pathology Non-GYN request.

### **RENAL PELVIS and URETERS**

Voided urine, retrograde catheterization, and direct brushings are satisfactory procedures for suspected lesions of the renal pelvis or the ureter and may assist in the localization of the lesion of the upper urinary tract.

Specimen must be labeled as to type (voided or catheterized), and for ureteral specimens and brushings, as to origin (right or left). Label and send specimen immediately to the Cytology Laboratory accompanied by a Cerner ordered: Pathology Non-GYN request.

### **TZANCK PREPARATIONS**

Multiple scrapings are taken from patients with localized herpes zoster, disseminated herpes zoster or herpes simplex infections.

The scrapings are prepared by selecting an early intact vesicle and swabbing the lesion with a swab saturated with 70% isopropyl alcohol. A blade or needle is used to open the vesicle and the lesion edge is gently scraped to avoid hemorrhage. The material is placed on clean, previously albumenized glass slides and gently spread.

Fix slides IMMEDIATELY by placing in 95% ethyl alcohol. Label and send specimen to the Cytology Laboratory accompanied by a Pathology requisition form.

Reference: Koss, LG, Diagnostic Cytology and Its Histopathologic Bases, 3rd Ed., Vol. 2, pp 1165-1187; 1979.