Directory of Pathology Services and Specimen Collection Manual

Introduction

General Information

Idaho Pathology Laboratory provides pathology services to Portneuf Medical Center, Pocatello, Idaho.

Portneuf Medical Center provides services in areas of surgical pathology, cytopathology, flow cytometry, hematopathology, forensics, immunohistochemistry, and molecular diagnostics. Portneuf Medical Center operates sections in surgical pathology, cytology (gyn and non-gyn), and histology.

Mission Statement

To improve the health and quality of life of the patients and communities we serve by providing the highest quality laboratory services by means of state-of-the-art technology and sound business practices.

Technology

The Surgical Pathology Laboratory and staff have access to a wide variety of ancillary techniques including immunohistochemistry, special stains, fluorescence immunopathology, flow cytometry, and molecular diagnostics. These ancillary services are performed to ensure optimal turnaround times for patient care.

- I. Leica Automated conventional tissue processor
- 2. Leica Bond machine for immunohistochemistry with approximately 40 antibodies
- 3. Digital cameras for macroscopic and microscopic pictures
- 4. Nova Path reporting system for surgical pathology and cytology reports

Pathology Client Services

Service Commitment Portneuf Medical Center believes in working closely with our clients to ensure quality patient care as well as client satisfaction. Our primary goals are to offer superior testing and personalized service through building professional relationships with our clients. Portneuf Medical Center's service commitment is to exceed the needs of its customers and support its clients in their own delivery of service.

Surgical Pathology

Introduction

The Surgical Pathology Laboratory offers general community services with advanced techniques in a wide variety of organ systems. All pathologists are certified by the American Board of Pathology.

Turnaround Time

- For routine specimens not requiring special techniques, a written report and telephone consult can usually be provided within one to two business days.
- For specimens where immunohistochemistry is required, a written report and telephone consult can usually be provided within three to four business days.
- For specimens where advanced diagnostic techniques are required (such as molecular studies), a written report and telephone consult can usually be provided within 7 to 10 business days.

Pathologists

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Contact Information

Pathology: 208-239-1690

FAX: 208-239-3711

PMC Laboratory: 208-239-1671

Specimen Handling Instructions

All specimens prepared for transport should follow these general instructions:

Diagnostic Specimen

A diagnostic specimen is defined as any human, including but not limited to: excreta, secreta, blood and its components, tissue and tissue fluids, being transported for diagnostic purposes. Specimens should be considered diagnostic specimens and sent under IATA Packing Instructions #650, unless the specimen is known to be an infectious substance.

Infectious Specimen

An infectious substance is defined as a viable microorganism, or its toxin, that causes or may cause disease in humans or animals, and includes those agents listed in 42 CFR 72.3 or the regulations of the Department of Health and Human Services and any other agent that causes or may cause severe, disabling, or fatal disease.

Specimens which are known or thought likely to contain infectious material must be packaged or sent separately following IATA Packing Instructions #602. Infectious specimen quantity must not exceed 50 mL.

Sending Specimens to Portneuf Medical Center

Specimen Transport Instructions

1. Complete a separate pathology request form for each patient (Histology or Cytology, as the case may be).

2. Be sure to record the following:

- a. Patient name, birth date, gender, and SSN
- b. Insurance and / or billing information including name of the insured, subscriber number, group number, name and address of insurance company and IPA group (if applicable)
- c. Site and type of biopsy
- d. Request for any special stains or studies
- e. Copy (front and back) of patient's insurance card
- f. Collection date and time
- g. Type of specimen
- h. Clinical history --- include a copy of prior pathology report if relevant
- 3. Mark box(es) indicating the test(s) requested
- 4. Print patient name and tissue source / location legibly on each specimen container (not on the lid)
- 5. If re-biopsy of a lesion, please provide date and the specimen accession number of previous biopsy.

A completed Surgical Pathology requisition with clinical history must be submitted with all cases. Please provide as much clinical history as possible, including history of prior malignancies. This information is important to the interpretation of the lesion and may help in selecting the appropriate special stains and tumor markers needed to make an accurate diagnosis.

Specimen Preservation

Biopsy specimens

1. Gross tissue specimens must be submitted in 10% neutral buffered formalin with enough fixative to cover the specimen. Specimen containers must be securely tightened to eliminate any leakage and must also display the OSHA required FORMALDEHYDE cautionary information.

2. For H & E slide consultations, specimen submission should include special stained slides, and / or paraffin blocks. Most special stains can be performed on conventional paraffin sections. Paraffin blocks are preferred for immunoperoxidase studies. If mercurial fixatives such as 85 are used, this should be designated on slides or blocks.

3. The following specimens should be substituted immediately WITHOUT FIXATIVE and may require special processing. A. Frozen sections B. Lymph nodes (place in RPMI solution)

4. It is best to confer with a pathologist concerning requests out of the ordinary prior to submission of the specimen.

Fluid specimens

1. Fix by mixing with equal amounts of 95% ethanol or 70% isopropyl alcohol (available in the hospital surgical area).

- A. Bladder washings
- B. Bronchial wash fluid and lavage fluid Do not add fixative. Refrigerate.
- C. Spinal fluid
- D. Pleural and ascetic fluid
- E. Synovial fluid
- F. Breast cyst fluid

Biopsy: Bone Marrow

Bone marrow biopsies are a sendout test to Genoptix Medical Laboratories. Handling should be done in accordance to Genoptix protocols.

Comment: Bone marrow biopsy specimens are performed for a variety of reasons ranging from evaluation of anemia to the diagnosis of metastatic carcinoma and leukemia. Complete patient histories, as well as documentation of any supporting details are vital to the complete and accurate interpretation of the material.

Routine Cervical, Endocervical, Endometrial Biopsies

1. Provide clinical history, i.e. last menstrual period, prior biopsies or pap smear information, and any history of hormone use including birth control pills on the requisition form.

2. Immediately place the specimen in the fixative container, tightly close container lid, and forward to the laboratory with the requisition.

3. Use of gauze pads to hold or place endometrial and endocervical sampling is discouraged because portions of the specimen are absorbed into the coarse weave and lost. Telfa is acceptable; placing the specimen directly into formalin is preferred.

Biopsy: Cone and Leep Conization of Cervix

1. Include on the requisition a history of prior pap smear or biopsy results.

2. If necessary, orient cone specimen with surgical suture material.

3. Immediately place the specimens in the fixative container, tightly close container lid and forward to the laboratory with the requisition.

Biopsy: Routine and Skin Excisions

1. Include any history of or gross description of lesion and history of previous biopsies if available,

- 2. Orient specimen as necessary using description or surgical suture material
- 3. Place the specimen in the fixative container and forward to the laboratory

Comment: Each biopsy should be placed in a separate container labeled with the site of biopsy. The consequences of submitting skin biopsies of several sites in a single container can be disastrous. Many dermatologic disease processes look similar microscopically. Please describe the lesion in detail, and provide as much clinical history as possible. Providing this information will help our pathologist to be very specific with his / her diagnosis.

Lymph Node for Flow Cytometry

1. Do <u>not</u> submit in formalin. Instead submit in RPMI fluid. Complete requisition and include primary care physician or oncologist.

2. Specimen should be received within 30 minutes for optimal results

Lymph nodes other than for flow cytometry should be submitted in 10% formalin for routine studies

Biopsy: Prosthetic Breast Implants

1. The prosthetic breast implants must be submitted in separate containers without fixation.

- 2. The fibrous tissue capsules may be submitted separately in 10% formalin.
- 3. Indicate on the requisition the presence or absence of prosthetic rupture or leakage.

Comment: Please indicate if these specimens are to be returned to you or the patient. A signed release form from the patient must accompany the request for return of specimen for medical legal purposes.

NOTICE: We have discovered that formalin fumes are extremely detrimental to cytology specimens. Many smears have been ruined because of exposure to formalin. This exposure often occurs during transportation when smears are shipped in the same container with biopsies.

We recommend shipping cytology and surgical specimens in separate specimen bags to minimize this potential problem.

For cell surface marker studies by flow cytometry, fresh tissue is desirable for improved preservation of antigens. The tissue should be placed in RPMI.

Arrangements for shipping and specimen containers can be made by contacting the pathology department at (208)239-1690.

Specimen Labeling Policy

To assure positive identification and optimum integrity of patient specimens from the time of collection until testing is completed and results reported, the client must label all specimens submitted for testing with the patient name and tissue source. Portneuf Medical Center samples from the same patient on the same day should also be labeled with the time of collection. Clients will be notified of inappropriately labeled specimens and the specimen will be returned to the client upon request.

Packing Requirements

1. Place the specimen in the zip lock portion of the specimen bag.

2. Fold completed requisition in half, and place the form in the outside sleeve of the specimen bag.

3. Ship one anatomic pathology specimen per bag.

Containers

To ensure safe handling procedures, non-compromised specimens, and to provide quality patient care, including fast and accurate test results, we request that Portneuf clients use the following guidelines

Acceptable Containers

-Portneuf Standardized containers

Unacceptable Containers/ Conditions

-The following will not be accepted because of safety and biological contamination issues:

- Leaky specimens that are not placed in a secured secondary container or leaking in such a way as to compromise testing
- Syringes with needles attached
- Transfer tubes secured with parafilm or paraffin

Hours of Operation

Regular work hours are 8:00 a.m. to 4:00 p.m., Monday through Friday. After hours; please see the pathology call schedule if in need of a pathologist.

Requisition Forms Histology (Surgical) Requisition Form

Cytopathology

Introduction

The Cytopathology Laboratory is a full-service laboratory, providing routine screening and diagnostic testing of all non-gynecologic, and fine-needle aspiration specimens. Our mission is to provide cytopathology service of the highest possible quality to clients and providers, as well as their patients.

Non-Gynecologic Cytology

The Cytopathology Laboratory provides non-gynecologic cytology services (including the evaluation of the following specimen types):

 Pulmonary: sputum, bronchial brush / wash and broncho-alveolar lavage (includes evaluation for opportunistic infections with Gram, AFB, and GMS stains; cell count and differential also available)
Urologic: urine (voided, catheterized, or instrumented), bladder wash, ureteral / renal pelvic wash / brush

3. Body Cavity Fluids: pleural, pericardial, pelvic wash, synovial

4. Cerebrospinal Fluid (includes cell count and differential for malignant CSFs for purposes of monitoring treatment)

5. Gastrointestinal: oral cavity / esophageal gastric brush / wash, bile, biliary system brush

6. Other Sources: nipple secretion, skin scraping (including Tzanck smears for detection of herpetic infection)

General Requirements

Fixation:

1. For cytological specimens, immediate fixation is essential for cytological examination. Air-drying begins as soon as the smear is made and proceeds rapidly although no grossly visible changes in the appearance of the smear or samples are present. Ethanol and methanol are acceptable alcohol fixatives; however, isopropanol is not acceptable. Stage your exam so that fixative can be applied immediately to the specimen using one of the following methods:

2. Fixative sprayed on smears immediately. Spray fixatives should be applied to smears with the spray can held 6 - 8 inches away from the slide.

3. Slides can be immediately placed in 95% alcohol fixative,

4. The cellular material can be immediately placed directly in 50% alcohol.

5. Please notify the pathology lab the type of fixative used, so the appropriate staining technique may be utilized.

6. Identify on the requisition the specimen site, method of sampling, (i.e. FNA, lump, lateral upper quadrant etc.).

Cytology: Induced Sputum and Bronchial Washings for Pneumocystis Pneumonia

Complete requisition and include request for "Pneumocystis jirovecii" pneumonia, (R/O PCP)
Enter washings into specimen container with precise identification of origin (i.e. right upper lobe of lung, etc.)

3. If brushings are performed concurrently, rotate brush gently over slide to apply material.

Comment: Specimens should be submitted in equal amounts of 50% ethanol, 70% isopropyl alcohol, or Saccamanno's cytology fixative. Only induced sputum or bronchial lavage / wash specimens are acceptable for ruling out PCP. Routine sputum for "rule out PCP" cannot be accepted.

Cytology: Bronchial, Colonic, Esophageal and Gastric Washings

1. Complete requisition as requested and include special stains if indicated.

2. Enter washings into specimen container with precise identification of site (i.e. right upper lobe of lung, etc.)

3. Add equal volumes of preservative fluid to each specimen. Close specimen container tightly. Do not add fixative. Refrigerate.

4. If brushings are performed concurrently, rotate brush gently over slide to apply material and fix immediately with spray.

Comment: Smears may be either air-dried, fixed with 95% alcohol, or spray fixed. If 95% alcohol or spray fixation is used, smears must be immediately fixed and designated as fixed on the requisition.

Cytology: Sputum

1. If specimen is collected in one single container, indicate this on the requisition form.

2. Have patient rinse mouth prior to collection.

3. Give container to patient and instruct to breathe deeply for 3 minutes.

4. Instruct patient to cough deeply from the diaphragm, with effort to expectorate material into collection container.

5. At short intervals, repeat the coughing attempts three more times with collection of all coughed up material.

6. Add equal volumes of fixative fluid and shake the container vigorously each time a new specimen is added to the cup (make sure that the container is tightly sealed before shaking).

7. Tightly close the collection container and forward to the laboratory with the completed requisitions form.

Comments:

1. Deep cough(s) from the diaphragm are necessary to provide an adequate specimen from the lower respiratory tract. The laboratory will determine adequacy of specimen by the presence of alveolar macrophages within the specimen. A series of three specimens in the morning on consecutive days are encouraged.

2. If there is a difficulty in producing a specimen, collection may be facilitated by the moisture and steam of a preceding, long hot shower. For patients unable to produce, consider aerosol induced coughing and specimen collection.

3. Sputum's collected post bronchoscope may be productive with diagnostic material even with negative bronchial washings, brushing, and biopsies.

4. Specimens consisting of saliva or nasal pharyngeal drainage will be reported as lacking alveolar macrophages, metaplastic cells or bronchial columnar cells. These specimens are inadequate for lesions of the lower respiratory tract and will not be considered as true negative studies.

5. With sputum samples positive for malignant cells, a primary of the head and neck region should be considered as well as malignancies of lung. Up to 10% of positive sputum's may reflect malignancies of the head and neck.

6. Specify the need for asbestos body examination. Studies will include examination of Papanicolaou and Prussian blue stained smears.

7. Specify the need for identification of pneumocystis carinii jirovecii. Specimens will include examinations of Papanicolaou, Diff Quick, and GMS as needed.

<u>Cytology: Cervical and Endocervical Smears (Pap Smears) are not processed at Portneuf</u> <u>Medical Center</u>

Pathology Cytology: Body Fluids

1. Materials: 50% methyl or ethyl alcohol, 70% isopropyl alcohol, or Saccomanno's cytology fixative.

Comments:

1. Body fluids to be processed in this manner include pleural fluid, ascetic fluid, cul-desac fluid and pericardial fluid.

2. The entire collected specimen should be submitted for cytological processing after aliquoting specimens for other studies (i.e. culturing, cell count, and protein analysis). It is our opinion that the yield and diagnostic sensitivity is increased when the entire specimen is received for cytological preparation and interpretation.

Cytology: Urine and Bladder Washings

1. Void and discard the first urine specimen. Patient should drink as much water as possible an hour prior to collecting the specimen.

2. Mix an equal amount of fixative with the urine specimen. Forward to laboratory with requisition form. **Comment:**

1. Specimen should be identified as sample type: (i.e. voided, catheterized, right or left ureteral or bladder irrigation fluid)

2. For detection of cancer in ureters or kidneys, a serial collection of specimens spaced over three days has proven to increase diagnostic sensitivity and yield.

3. Unpreserved urine results in the rapid degeneration of exfoliated cells.

Cytology: Nipple Secretions, Smears, Brushings

1. Include pertinent history and site of sample obtained on requisition.

2. If there is no nipple erosion or ulceration, gently "strip" the area of the breast below the nipple and areola with a motion form beneath the areola towards the nipple surface. Do not massage the breast. With the appearance of fluid, touch a slide to the drop of fluid and draw the slide quickly across the nipple. Repeat the process for opposite breast.

3. If there is nipple erosion or ulceration, touch a slide to this area three times, with a different part of the slide.

4. IF no fluid can be expressed, a swab may be dipped in saline and gently rolled and rotated on the ulcerated surface, and applied to a glass slide.

Comment: The obtained smears can either be completely air-dried or spray fixed. If spray fixation is elected, it is vitally important that smears be sprayed as soon as obtained. Partial fixation of slides develops air drying artifact that will only complicate interpretation and may even result in an unsatisfactory specimen. It is important to indicate the type of fixation (air dry vs. fixed smears) so that the proper staining procedure may be selected at the laboratory. We recommend completely air drying smears to avoid fixation artifact.

Fine Needle Aspiration

Portneuf Medical Center:

Fine needle aspiration provides a prompt, cost effective, safe, simple and useful evaluation of a mass through cytological diagnosis that is usually well tolerated by patients. It is often used as an alternative to surgery and may provide a definitive diagnosis that will determine therapy and / or assist in a planned surgical approach with effective utilization of operating room time. In general, any palpable mass can be evaluated by aspiration techniques. With ultrasound guidance, fluoroscopy, and CT, most deep-seated lesions may also be sampled. Lesions that are commonly sampled include: thyroid, breast, salivary glands, and lymph nodes.

Materials Required:

1. Frosted end labeled slides.

2. Specimen containers with appropriate fixative as needed (Saccomanno's fixative , or 50% ethyl or methyl alcohol) .

- 3. Spray fixative.
- 4. Requisition form. The obtained smears

Procedure:

1. Label slides and specimen containers with the patient's name and site of aspiration.

2. Place labeled slides in three rows of four slides, each row for use with a "pass". This set-up supports three passes. Open container with fixative for needle washings.

3. Place the bevel of the needle against a glass slide and express a small drop of aspirated material onto the slide.

a) If too much material is expressed onto the slide, either re-aspirate a portion of the material by withdrawing the syringe plunger slightly, or spread the material out among several slides.

b) If the cellular material is semi-solid, place a second slide on top of the material and pull the slides gently and quickly apart as the material spreads from the weight of the slide.

c) If the aspirated material is diluted by fluid or by blood, use the same smear technique as for blood smears:

I. Back the edge of a slide or coverslip into the drop and as the material spreads along the edge, move the slide / coverslip forward pulling the cellular material away from the fluid or blood.

II. Immediately fix the smear for Papanicolaou stain with spray fixative or 95% ethyl or methyl alcohol. Or alternatively:

III. Allow smears to air-dry for Diff Quick staining. Write "air dried" on the end of the smears.

IV. If staining is available, have the patient remain while adequacy of the aspiration pass is determined and repeat the procedure until the operator is satisfied with the adequacy of the material.

V. Label all slides with the patient's name and site of aspiration.

d) Complete requisition form, including patient name, birth date, date of service, and billing data.

Immunohistochemistry

Introduction

The Immunohistochemistry Laboratory has expertise in all aspects of diagnostic immunohistopathology. The section performs immunohistochemistry using antibodies directed against a wide variety of antigens. Both individual antibodies and diagnostic panels of antibodies can be ordered through this section. The expertise of this section ensures that the studies are performed and interpreted to a high standard of technical excellence. Consultation services are available to help determine the most appropriate individual test or panel of tests to be run for specific differential diagnostic problems.

- The Immunohistochemistry Laboratory at Portneuf Medical Center provides the following services: Lymphoma and Leukemia Phenotyping Undifferentiated Tumor Phenotyping
- Differential Diagnosis Analysis of metastasis of unknown primary (TLLP)
- Lymphoma vs. Carcinoma Adenocarcinoma vs. Mesothelioma Melanoma vs. Carcinoma vs. Lymphoma Hodgkin's vs. Non-Hodgkin's Lymphoma
- Cell Lineage and Tumor Primary Site Determination Detection of Infectious Agents

Paraffin-Embedded Tissue

•Collect: Tumor tissue.

•Transport: Fixed, paraffin embedded tissue is preferred. If the tissue block cannot be sent, send sections mounted on 2% aminoalkylsilane-treated slides or positive (+) charged slides. Paraffin blocks and slides must be adequately protected to prevent melting in the summer months by shipping blocks in cooled containers.

Fresh Tissue

•Specimen must reach the Hematopathology Laboratory within 24 hours of collection to avoid significant deterioration.

•Collect: Tumor tissue.

•Transport: Fresh tissue wrapped in gauze dampened with normal saline or PBS, place in a sealed container, and transport on a generous amount of "wet ice." Use refrigerant pack in place of "wet ice", if transport time is less than one hour.

Peripheral Blood and Bone Marrow

•Samples must be received within 24 hours of collection and kept at room temperature to be viable for evaluation.

•Collect: Peripheral blood and bone marrow in EDTA (purple) or Heparin (green) tubes.

•Transport: Anticoagulated blood and smears at room temperature to Portneuf Pathology.

•Note: Blood, bone marrow smears, and cytocentrifuged preparations should first be air dried and sent unfixed. Slides should be sent at room temperature and protected from light exposure.

Billing Procedures

Billing charges will be assigned after the testing has been completed. Patients will be billed only for the services they receive, which will be a variable price based upon the testing performed.

You will receive two billing statements: Idaho Pathology Laboratory will bill for the professional interpretation of the bill and you will receive a second bill from Portneuf Medical Center for the technical portion of the diagnostic specimen.

The Surgical Pathology and Histology Laboratory is dedicated to the production of highly technical, quality sections and a wide range of expertly performed special stains. The laboratory will accept formalin fixed tissue or paraffin blocks and pre-cut slides for special staining techniques.

Special Staining Techniques

Portneuf Medical Center offers a complete directory of routine special staining techniques:

AFB (Ziehl-Neelsen Method)

Iron

Alician Blue pH 2.5

Grocott's Methenamine Silver Method (GMS)

Iron Stain (Mallory's Method)

Periodic Acid Schiff (PAS)

PAS with Diastase Digestion

Gill Acid-Hematoxylin 3

Reticulum Stain

Gamori's Trichrome

Elastic Stain

Dif Quik

Hematoxylin and Eosin

Updated 4/15/2022 KTV