



Laboratory Medicine

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Cytology Collection Manual

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1.0 Contact Information:
EASTERN HEALTH
CLINICAL CYTOPATHOLOGY
DIVISION OF LABORATORY MEDICINE

Laboratory: Phone: (709) 777-6380/6381
Fax: (709) 777-8583

Clinical Chief: Ed Randell Dr.
(709) 777-6921

Director: Dr. C Ghosh
(709)777-2108

Operations Manager: Trevor Williams
(709) 777-2275

Location: Cytology Laboratory
3rd Floor Southcott Hall
L.A. Miller Centre
100 Forest Road
St. John's, NL
A1A 1E5

Hours: 0800 - 1600 hours, Monday to Friday

2.0 General Information:

2.1 Urgent Requests:

It is the Health Care Provider's responsibility to notify the Cytology Laboratory of an urgent request, in accordance with OP-CYT(PCO)-005 Submission of Urgent Cytology Specimens. The Health Care Provider must provide their contact information to the laboratory so urgent results can be communicated to them in a timely manner.

2.2 Processing and Hours of Operation:

Specimens are not routinely processed after 1600 hours. Every effort should be made to submit specimens during laboratory hours. Specimens should be fixed to ensure optimum conditions for microscopic examination. Specimens received after hours will be processed the next working day.

2.3 Submission of Specimens:

Identification of the specimen should be placed on the body of the container, **not** on the lid.

DO NOT send fluids in syringes (with or without needle attached).

DO NOT send fluids in collection or catheter bags.

DO NOT send large volumes of fluid.

2.4 Fixative Storage, Stability and Precautions:

CYTORICH® Red Preservative Fluid:

- Should be stored at room temperature (15-30C)
- Specimens fixed in CYTORICH® Red Preservative Fluid are stable for at least 30 days.

- Do not use CYTORICH® Red Preservative Fluid beyond the expiration date posted on the specimen container.
- Specimens fixed in CYTORICH® Red Preservative Fluid can withstand shipment at temperatures from about 2° to 30°C.
- Do not allow reagent to contact open wounds.
- Wear powder-free gloves and eye protection. Avoid splashing and generating aerosols.
- Do not ingest (contains alcohols other than ethanol and formaldehyde).

Sure Path™ Preservative Fluid:

- Storage condition of fluid without cytologic specimens is up to 36 months from date of manufacture at room temperature (15° to 30° C)
- Storage limit for fluid with cytologic specimens is 6 months at refrigerated temperatures (2° to 10° C) or 4 weeks at room temperature (15° to 30° C).
- Sure Path™ Preservative Fluid was tested for antimicrobial effectiveness however universal precautions for handling should be practiced.
- Avoid splashing or generating aerosols; use appropriate hand, eye, and clothing protection
- The mixture contains small amounts of methanol and isopropanol. Do not ingest.

FIRST AID for CYTORICH® Red Preservative Fluid and Sure Path™ Preservative Fluid:

- Inhalation: Remove to fresh air and rest
If recovery is not rapid call for prompt medical attention
- Eyes: Irrigate with water for at least 15 minutes
Take care not to wash chemical from one eye to another
If irritation persists, obtain medical attention
- Skin: Remove contaminated clothing
Wash with soap/cleanser and rinse with plenty of water
If irritation persists, obtain medical attention
- Ingestion: Do not induce vomiting
Give plenty of water to drink
Beware of aspiration if vomiting occurs
Seek medical attention immediately

2.5 Transport of Specimens to the Cytology Laboratory:

1. Specimens must be submitted in a specimen container with an alcohol-based fixative, such as Cytorich Red.

2. The specimen container, as well as the requisition, must contain the appropriate patient identifiers: date of collection, name of patient, MCP number, and specimen type.
3. The specimen container must be sealed in an appropriately sized biohazard specimen bag with the requisition in the outside pouch.
4. The biohazard bags containing the specimens must be transported in opaque, hard walled courier bags that have been sealed with zip tags. Do not use plastic or paper bags to transport cytology specimens to the laboratory.
5. A transportation log must be contained within the courier bag itemizing each specimen being sent.
6. Extremes in temperature during transportation must be avoided. The acceptable temperature range is 4°C – 30°.
7. Deliver to the laboratory in a timely fashion after collection. Specimens older than 35 days are not suitable for HPV testing.

3.0 Collection of Non-Gynecological Specimens for Cytology:

3.1 Specimen Types:

Respiratory samples including but not limited to:

- Bronchial washings – right upper lobe (**RUL**), right middle lobe (**RML**), right lower lobe (**RLL**), left upper lobe (**LUL**) and left lower lobe (**LLL**)
- Bronchioloalveolar Lavage - right upper lobe (**RUL**), right middle lobe (**RML**), right lower lobe (**RLL**), left upper lobe (**LUL**) and left lower lobe (**LLL**)
- Sputum – Expecterated, Post Bronchoscopy, Induced/tracheal aspirate
- Bronchial brushing - right upper lobe (**RUL**), right middle lobe (**RML**), right lower lobe (**RLL**), left upper lobe (**LUL**) and left lower lobe (**LLL**)

Gastrointestinal samples including but not limited to

- Esophageal brushings
- Gastric brushings
- Common Bile Duct brushings
- Oral scrapings
- Other miscellaneous brushings
- Gastric Washings

Body cavity fluids including but not limited to:

- Cerebral Spinal Fluid - CSF
- Pleural Fluid – right or left
- Pelvic Washings (including Diaphragm Washings, Gutter Washings, Cul De Sac Washings)
- Peritoneal Fluid
- Pericardial Fluid
- Synovial Fluid
- Ascites Fluid
- Nipple Discharge
- Other Miscellaneous Body Cavity Fluids

Urinary tract samples including but not limited to:

- Voided Urine
- Catheter urine
- Cystoscopic Samples (Bladder Washings, Renal Pelvic Washings/ Brushings, Ureteral Washings/ Brushings/Ileal Conduit)

3.2 Non-Gynecological Specimen Collection

Verify the identification of the patient in accordance with the Positive Patient Identification Policy (PRC-130).

1. **Cytology studies** – Specimen should be fixed using CytoRich Red fixative – if no CytoRich Red Fixative is available 50% alcohol must be used.
 - Ideally specimens submitted must be fixed using at least equal volume of fixative to specimen, however it is acceptable to have more fixative than specimen. **(Ideally a 1:1 ratio.)**
 - If there is a visible clot and/or tissue fragment place the clot or tissue fragment in 10% Neutral Buffered formalin
2. **For PD-L1 and/or Molecular Studies** – Specimen must be fixed using 10% Neutral Buffered formalin.
 - Ideally specimens submitted must be fixed using equal volume of fixative to specimen, however it is acceptable to have more fixative than sample
3. **If there is suspicion of hematological malignancy(Lymphoma or Leukemia)** - Specimen must be collected in Flow Cytometry collection media – Flow media in addition to cytology fixative

4. OIL RED O Stain for fat: - Specimen must be collected fresh**Important Notes:**

- If no fixative is available, refrigerate fresh specimen at 4 degrees Celsius for a period of less than 24 hours. Deliver to immediately to the Cytology Laboratory.
- Leaking containers pose potential hazard and cannot be processed and will be discarded in accordance with 410-QP (PCO)-004 Specimen Acceptance and Rejection
- In case of large volume of fluid sample ensure that specimen is thoroughly mixed before placing specimen in fixative
- Specimens are to be brought to lab office located at Health Sciences Center or lab office located at St. Clare's Hospital
- When specimen is collected on the weekend, please store sample in the refrigerator at 4 degrees Celsius

3.3 Requisition Form and Sample container:**All specimens collected must have accompanying requisitions completed.**

- Number 1 and 2 above must have a Cytology Non Gyn Requisition completed and number 3 must have a Flow Cytometry Requisition completed. All information on specimen container and requisition must match. Any discrepancies will cause delay in specimen acceptance and specimen processing.

The following **must** be included on the requisition and sample container:

- **Patient's name** – must be clearly marked on sample container and requisition
- **Health Insurance number** (MCP#) - must be clearly marked on sample container and requisition
- **Source of specimen** - must be clearly marked on sample container and requisition
- **Date and time of collection** - must be clearly marked on sample container and requisition

The following **must** be included on the requisition:

- **Full Clinical History** – clearly mark on requisition (including type of cancer)

- **Request for Biomarkers/Molecular testing-** clearly marked on requisition
- **Request for OIL RED O Stain for fat** must be clearly marked on requisition

Note: An interpretative diagnostic report will be sent to the submitting Health Care Provider

4.0 Collection of a Fine Needle Aspiration for Cytology:

Fine needle aspiration is applied to palpable lesions or deep-seated lesions under guided imaging techniques such as Computed Tomography CT/ Ultrasound, Endoscopy, Bronchoscopy, EBUS guided, and WANG needle procedures.

The lesion must be sampled by directing the needle into the target lesion. Multiple passes increase the diagnostic sensitivity.

4.1 Specimen Types:

Respiratory specimens including but not limited to:

- Lung - right upper lobe (RUL), right middle lobe (RML), right lower lobe (RLL), left upper lobe (LUL) and left lower lobe (LLL)

Lymph node specimens including but not limited to:

- Intra-thoracic lymph nodes, mediastinal lymph nodes and hilar lymph nodes
- Lymph nodes associated with the head and neck – submandibular, cervical, intraparotid etc.
- Inguinal lymph nodes
- Retroperitoneal lymph nodes
- Axillary lymph nodes - left or right

Head and Neck specimens including but not limited to:

- Thyroid gland – right, left or isthmus
- Parotid glands – right or left
- Salivary glands – right or left submandibular, left or right sublingual gland
- Thyroglossal duct - right or left
- Other – Brachial cleft cysts

Other anatomical sites including but not limited to:

- Breast – right or left

- Liver
- Pancreas
- Omentum
- Female genital tract - ovaries (left or right)
- Bone – sacrum, ribs, etc.
- Soft tissue lesions – lipoma, melanoma, etc.

4.2 Fine Needle Aspiration Specimen Collection

Verify the identification of the patient in accordance with the Positive Patient Identification Policy (PRC-130).

Specimens should be preserved in the appropriate fixative immediately upon collection – specimen should be collected as follows:

- 1 Cytology studies** – Specimen should be fixed using CytoRich Red fixative – if no CytoRich Red Fixative is available 50% alcohol must be used.
 - Ideally specimens submitted must be fixed using at least equal volume of fixative to specimen, however it is acceptable to have more fixative than specimen. **(Ideally a 1:1 ratio.)**
 - If there is a visible clot and/or tissue fragment place the clot or tissue fragment in 10% Neutral Buffered formalin
- 2 For PD-L1 and/or Molecular Studies** – Specimen must be fixed using 10% Neutral Buffered formalin.
 - Ideally specimens submitted must be fixed using equal volume of fixative to specimen, however it is acceptable to have more fixative than sample
- 3. If there is suspicion of hematological malignancy (Lymphoma or Leu -**
Specimen must be collected in Flow Cytometry collection media – Flow media in addition to cytology fixative

Important Notes:

- If no fixative is available, refrigerate fresh specimen at 4 degrees Celsius for a period of less than 24 hours. Deliver to immediately to the Cytology Laboratory.
- Leaking containers pose potential hazard and cannot be processed and will be discarded in accordance with 410-QP (PCO)-004 Specimen Acceptance and Rejection

- Specimens are to be brought to lab office located at Health Sciences Center or lab office located at St. Clare's Hospital
- When specimen is collected on the weekend, please store sample in the refrigerator at 4 degrees Celsius

4.3 Requisition Form and Sample container:

All specimens collected must have accompanying requisitions completed.

- Number 1 and 2 above must have a Cytology Non Gyn Requisition completed and number 3 must have a Flow Cytometry Requisition completed. All information on specimen container and requisition must match. Any discrepancies will cause delay in specimen acceptance and specimen processing.

The following must be included on the requisition and sample container:

- **Patient's name** – must be clearly marked on sample container and requisition
- **Health Insurance number (MCP#)** - must be clearly marked on sample container and requisition
- **Source of specimen** - must be clearly marked on sample container and requisition
- **Date and time of collection** - must be clearly marked on sample container and requisition

The following must be included on the requisition:

- **Full Clinical History** – clearly mark on requisition (including type of cancer)
- **Request for Biomarkers/Molecular testing**- clearly marked on requisition

Note: An interpretative diagnostic report will be sent to the submitting Health Care Provider.

5.0 Collection of Liquid Based Pap for Cytology:

5.1 Specimen Type:

- Liquid-based Pap

5.2 Requisition Form:

- Record full name and address of the submitting Health Care Provider
- Record patient's full name, healthcare number and date of collection

- Gynecological Cytology requisition including relevant clinical history- **abnormal history**
- Source of specimen must be included – cervical/endocervical or vaginal
- Last menstrual period must be included – **LMP**

5.3 Specimen Collection Vial:

- Label the collection vial with patient's full name and healthcare number.

5.4 Supplies:

- SurePath™ Preservative Fluid Collection Vial (includes SurePath™ Preservative Fluid)
- Cervical Sampling Device(s) with detachable Head(s) – broom-type device or brush/plastic spatula.
- Speculum

LBC Pap Test Kits may be ordered by contacting Newfoundland Public Health Laboratories @ (709) 777 7242 or online @ www.publichealthlab.com

5.5 Specimen Collection:

Verify the identification of the patient in accordance with the Positive Patient Identification Policy (PRC-130).

- A mid-cycle smear is optimal
 - Sampling of the transformation zone of cervix is important for specimen adequacy. The presence or absence of transformation zone is reported in "Specimen Adequacy" in pre-menopausal women
 - Smears are not recommended during menstruation exception **abnormal bleeding**
1. After collection, break off tip of sampling device in the collection vial. Secure lid.
 2. Label vial with patient's full name and healthcare number. Place in a biohazard transport bag with completed requisition. Transport to the Cytology Laboratory.

For additional information on who is to be screened and Pap collection, refer to Cervical Screening Initiatives: Cervical Screening Guidelines for Newfoundland and Labrador

<http://www.centralhealth.nl.ca/assets/Department-Information/CSI-Files/Cervical-Screening-Guidelines-2011.pdf>

5.6 Preservative:

- SurePath™ Preservative Fluid (in the collective vial). Stable up to 36 months from date of manufacture at room temperature. Do not use beyond the expiration date posted on the vial.

5.7 Special Handling/ Transport:

- Ensure lids are secure before transport.
- Time restrictions impact testing for HPV; deliver to laboratory in a timely fashion after collection

Note: An interpretative diagnostic report will be sent to the submitting Health Care Provider

Appendix A – Quick Reference Pap Smear Requisition Completion

QUICK REFERENCE

Cytology Pap Smear Requisition



Affix Cytology
Laboratory Label
Here

PAP Smear Requisition

HCN: 000111222333 Prov/Terr: NL Expiry: 2020 / JAN / 01 Full Legal Name: JANE First ANN Middle DOE Surname DOB: 1980 / MAR / 10 Sex: ☐ M ☒ F ☐ UN Mailing Address: 1 COWAN AVE. City: ST. JOHN'S Prov: NL Postal Code: A1A1A1 Telephone: (Check Best) ☒ Home (709) - 111 - 2222 ☐ Cell () - () - () ☐ Work () - () - ()	
Ordering Provider's Name: DR. RICK WALSH Clinic Name: MEDICAL PRACTICE Mailing Address: 10 TOPSAIL RD. City: ST. JOHN'S Prov: NL Postal Code: A1A1A1 Phone: (709) - 999 - 9999 Fax: (709) - 999 - 9998 Ordering Provider's Meditech Mnemonic: WALRI Signature: <i>[Signature]</i> Date: 2021 / JAN / 06	Clinic Stamp: (include fax, provider and mnemonics) EMR Clinic Mnemonic: BROWJ COPY TO PROVIDER DR. JOHN BROWN
Date Specimen Taken: 21 / 10 / 02 Clinical History: Last Menstrual Period: 21 / 01 / 02 Abnormal Bleeding? ☐ Yes ☒ No Post-Menopausal? ☐ Yes ☒ No Previous Abnormal Papi/Colposcopy? ☐ Yes ☒ No HPV Vaccination? ☒ Yes ☐ No Date of Last Pap Smear: 18 / 03 / 01	Source of Specimen: ☒ Cervical/Endocervical ☐ Vaginal Medications: ☐ BCP ☐ HRT ☐ Chemotherapy/Radiation ☐ Depo-Provera ☒ IUD Other Significant History/Colposcopic Findings: N/A
Laboratory Use Only Lab Accession Number: _____ Date Specimen Received: 21 / 10 / 02	
Specimen Adequacy: ☐ Satisfactory for evaluation ☐ TZ Present ☐ TZ Absent ☐ Unsatisfactory ☐ No brush present ☐ Rejected	Adequacy Qualifier: ☐ Blood ☐ Bacteria ☐ Scanty ☐ Leaked Vial ☐ Inflammation ☐ Other ☐ Improper Labelling
☐ Negative (NILM) ☐ Shift in Flora ☐ ASCUS ☐ AGC-NOS ☐ Reactive ☐ Actinomyces ☐ ASC-H ☐ AGC-FN ☐ Inflammation ☐ Herpes ☐ LSIL ☐ AEC ☐ Trichomonas ☐ Rad/Chemo ☐ HSIL ☐ AEM C ☐ Yeast/Fungal ☐ Endometrial cells > 45 years ☐ SCC ☐ AIS ☐ Adeno ☐ CMV ☐ Other ☐ Other Malignancy	
Recommendations: ☐ Routine Screening ☐ Repeat 3 months ☐ Repeat 6 months ☐ Reflex HPV ☐ Colposcopy ☐ Biopsy ☐ ECC ☐ EM Sampling	
Technologist/Pathologist Signature: _____ Date: ____ / ____ / ____	

All shaded areas must be completed

It is critical to include the first and last names of all physicians or NPs named on the requisition to avoid delay or error in return reports. **Do not use initials. LABELS or STAMPS ARE ACCEPTABLE**

Requisition and Specimen Bottle must have 2 patient identifiers. Use same identifiers on both requisition and specimen. First must be the patient's full name, no initials, second may be:

1. MCP (preferred)
2. Date of Birth
3. Chart number
4. Other Health Card Number

LABELS ARE ACCEPTABLE



Use only ink pen or a label as felt marker rubs off specimen bottles. Specimen Bottle label must include 2 patient identifiers and date of collection – HPV testing can only be done on specimens <36 days old.



DO NOT USE PARAFILM TO SEAL SAMPLES, THIS CREATES LEAKAGE

Appendix B – Quick Reference Cytology Collection Instructions

QUICK REFERENCE

Cytology Collection Instructions

IMMEDIATELY FIX all Cytology specimens after collection.

Note: **OIL RED O requests for fat must not be fixed; specimen must be submitted fresh**

Label Cytology specimens and Cytology requisitions with TWO Patient Identifiers, the Source of the Specimen and Anatomic Site of Origin and the Date of Collection. Include clinical history and name of the submitting Healthcare Provider.

CytoRich® Red Preservative Fluid is the fixative of choice for Cytology **non-gynecological** specimens. Check name of fixative and expiry date. Fixative is available in specimen containers containing 25 ml. Under extenuating circumstances, when **CytoRich® Red Preservative Fluid** is not available, 50% alcohol may be used as an alternative preservative.

SurePath® Preservative Fluid is the fixative of choice for Cytology **gynecological specimens** (PAP). Check expiry date. Fixative is available in specimen vials with predetermined volumes.

DO NOT USE FORMALIN

SPECIMEN TYPE	FIXATIVE VOLUME	RATIO	SPECIAL HANDLING
Body Fluids (pelvic, urine, pericardial, pleural, sputum, peritoneal/ascites, cystic fluid, bronchial & gastric washings, synovial, other body fluids) Note: OIL RED O fat stain	CytoRich® 25 ml NO Fixative	1:1	Mix large volumes of fluid before taking a representative sample. No fixative – sample must be fresh
Fine Needle Aspirations	CytoRich® 25 ml container	Entire sample	Cyst aspirations refer to: Body Fluids.
Brushings (bronchial, gastrointestinal, common bile duct, miscellaneous brushings, oral scrapings)	CytoRich® 25 ml container	Brush tip Add oral scrapings to fixative	When submitting a brush, swish the brush in the fixative to release the material; cut brush tip off and leave in fixative.
Gynecological Specimens (PAP)	SurePath® Predetermined volume	Collection device head	Failure to submit the collection device head will represent a technical inadequacy and will be rejected.

INQUIRIES CONTACT: Cytology Laboratory, L.A. Miller Centre

Phone (709) 777-7926/6380/6381 FAX (709) 777- 8583 Business Hours: 0800–1600 Monday to Friday

ENSURE CAP IS SECURELY FASTENED ON SPECIMEN CONTAINER BEFORE TRANSPORT.