

**Atlantic Provinces Pediatric Hematology Oncology Network**

**Réseau d’Oncologie et Hématologie Pédiatrique des Provinces Atlantiques**

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*Reviewed and approved by specialists at the IWK Health Centre, Halifax, NS*

*and The Janeway Children’s Health and Rehabilitation Centre, St. John’s, NL*

**Guidelines for the Handling & Processing of**

**Potentially Malignant Specimens from Pediatric Patients**

*APPHON/ROHPPA supportive care guidelines have been developed by appropriate Atlantic Provinces health professional specialists (physicians, pharmacists, nurses and other health professionals) using evidence-based or best practice references. Format and content of the guidelines will change as they are reviewed and revised on a periodic basis. Care has been taken to ensure accuracy of the information. However, any physician or health professional using these guidelines will be responsible for verifying doses and administering medications and care according to their own institutional formularies and policies and acceptable standards of care.*

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**1. Purpose:**

To ensure appropriate handling and processing of **malignant or potentially malignant** pathology specimens from pediatric patients at institutions other than pediatric tertiary care centres.

**2. Rationale:**

There are important differences when handling pediatric specimens in comparison to the standard practices for adult cancer specimens.

The initial handling and processing of pediatric specimens (diagnostic biopsies or primary resections) is crucial to ensure collection of fresh tissue samples for diagnostic, prognostic, and predictive testing; and/or enrollment in clinical trials.

**3. Process:**

Whenever possible, if malignancy is in the differential diagnosis of a mass lesion in a child or adolescent, **referral to a tertiary-care children’s hospital is advised**. Pediatric surgeons and oncologists can help arrange timely work-up and biopsy.

If a biopsy procedure is to be carried out at a site without pediatric pathology services, consultation between local surgeon and/or pathologist with a pediatric pathologist is strongly recommended to discuss appropriate handling of the specimen. If the local pathology laboratory cannot provide appropriate tissue handling of potentially malignant specimens, then surgery should be referred to a site with pediatric pathology services.

Local pathologists should be aware of standard tissue handling recommended by and carried out in pediatric pathology laboratories. In particular, fresh **sampling** of lesional tissue for frozen storage is required.

**Specific specimen handling instructions:**

1. Specimen must be sent fresh/dry (**NOT in formalin or any other fixative**) to the pathology laboratory from the operating room, with cold ischemic time kept to a minimum (less than 15 minutes).

a. The laboratory must ensure timely receipt and handling of the specimen by a pathologist or appropriately supervised delegate.

2. Touch prep and/or frozen section may be considered by the pathologist to help guide specimen triage or to determine adequacy of sample. Frozen sections may be considered if they will influence intra-operative decisions.

3. A portion of the fresh lesional tissue **must be frozen** in a cryovial or another clean sealed receptacle.

a. Adjunct methods like liquid nitrogen can be used to aid flash-freezing but are not required.

b. Labelled specimen should be placed immediately in a negative 70oC to negative 80oC freezer.

c. If ultra-low temperature freezer is not available, specimen should be placed on dry ice or in a negative 20oC freezer until it can be transferred to colder storage as soon as possible.

d. The amount of tissue reserved for freezing depends on total quantity of lesional tissue available. For example, if multiple core biopsies are received, one entire core should be reserved for freezing. For larger specimens, multiple 0.5 to 1 g aliquots can be stored in separate cryovials. Even if lesional tissue is extremely limited, every effort should be made to save at least one small sample frozen.

4. **Optional** sampling of fresh tissue can be considered, depending on specific case, differential diagnosis, and quantity.

a. Microbiology cultures: should be considered for all lymph node specimens or whenever infection is in the differential diagnosis; bacterial, mycobacterial, and/or fungal cultures can be performed on fresh sterile samples per local laboratory protocols (consult with microbiology lab for special circumstances).

b. Cytogenetics (karyotype and/or FISH): karyotype no longer ordered routinely, but preservation of some fresh tissue in RPMI or other culture media may provide options for diagnostic FISH in some cases.

c. Flow cytometry: should be considered for all potential lymphomas; handle per local laboratory’s protocols.

d. Electron microscopy: no longer used routinely; a very small sample (1-2 mm3) can be fixed in glutaraldehyde if available.

5. After procuring mandatory sample for freezing the priority is for adequate tissue for microscopic work-up. Tissue can be transferred to an adequate volume of **formalin for fixation for routine processing** and paraffin embedding according to local laboratory standards. Ideal fixation time for most specimens is 24-48 hours.

a. It is advisable to divide lesional tissue into multiple blocks in case an entire block is required for diagnostic testing or donation to clinical trials group.

Questions can be referred to:

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