41 Lymph Nodes

High-quality sections for routine light microscopy are necessary, but not always sufficient, for the interpretation of lymph node biopsies. Immunophenotypic and genetic studies are often required for the diagnosis and classification of a hematopoietic neoplasm. Adequate fixation and timely and appropriate technical handling of lymph nodes are, therefore, even more important than with other specimens.

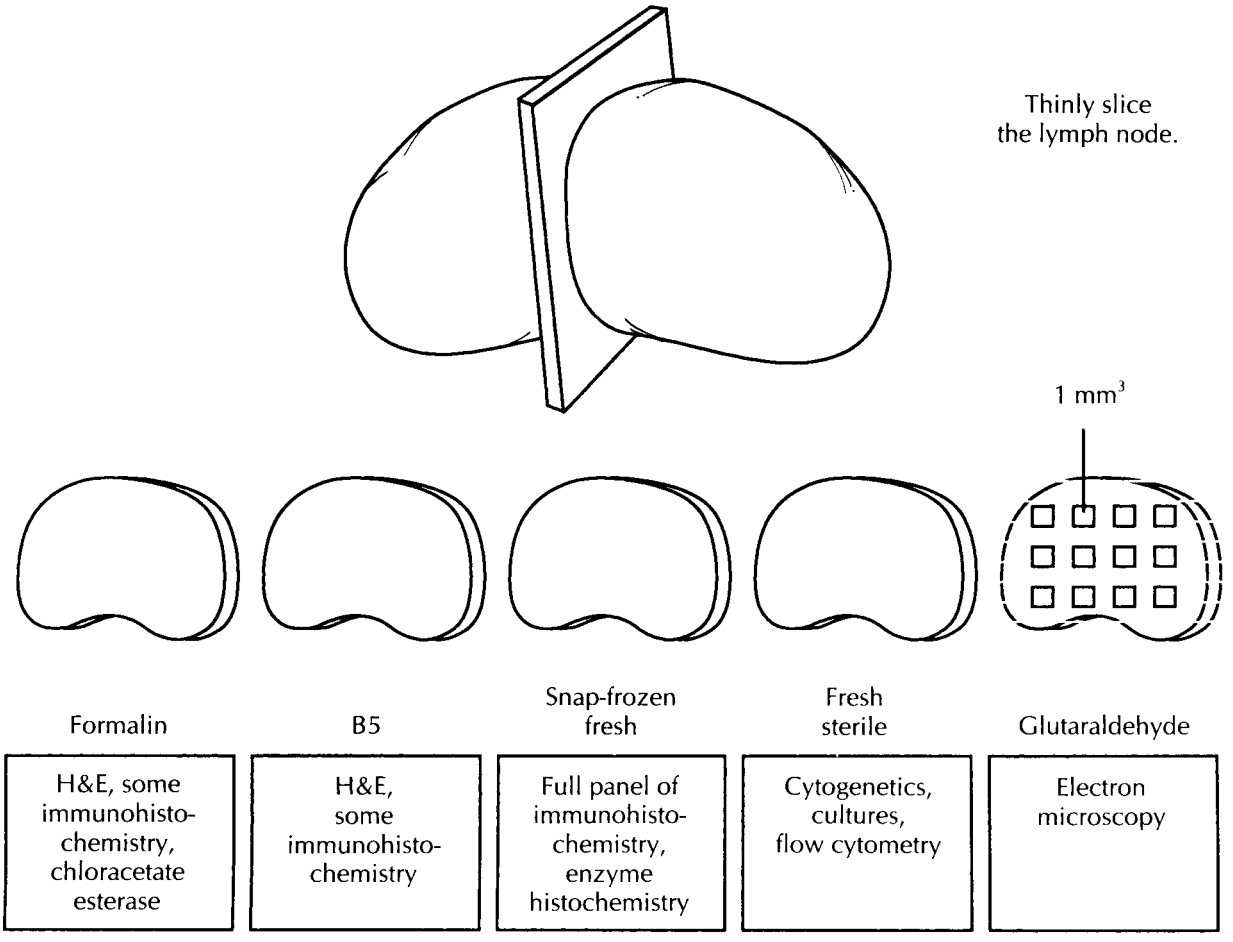
When lymph nodes are placed in an empty specimen container or in dry gauze, the edges of the specimen dry out, producing a prominent desiccation artifact at the edge of the node. Severe edge artifacts can be introduced into a lymph node even before the specimen reaches the surgical pathology laboratory. Surgeons should therefore be instructed to place resected lymph nodes immediately into a balanced physiologic solution such as Roswell Park Memorial Institute medium (RPMI) 640 or isotonic saline, and to transport lymph nodes immediately to the surgical pathology laboratory. Remember that lymph nodes can also dry out on the cutting table, so proceed quickly and efficiently after removing the specimen from the transport media.

Once the specimen is received, document its size, weight, and shape, and then slice it into uniformly thin 2- to 3-mm sections. Examine the cut surfaces of the node, and ask the following questions: Is the nodal architecture preserved? If the architecture is ablated, is the node grossly nodular, or is the process diffuse? Are any focal lesions present? Is the capsule intact? What is the appearance of the perinodal tissues?

Next, prepare touch imprints by placing the surface of a glass slide against the cut surface of the lymph node. At least five air-dried slides should be prepared, especially in cases of suspected Burkitt's lymphoma, lymphoblastic lymphoma, and myelogenous leukemia. These can be used later for Giemsa stains, oil red O stains, acid phosphatase stains, chloracetate esterase stains, and immunofluorescence for nuclear terminal transferase. Two additional imprints immediately fixed in 95% alcohol should be prepared for possible hematoxylin and eosin (H&E) staining.

Next, tissue should be submitted for light microscopy and, if sufficient tissue is available, for immunohistochemical and genetic studies. Sections for light microscopy should include not only the substance of the node, but also the capsule and perinodal soft tissues. Submit at least one section for fixation in neutral buffered formalin and at least one section in B-5 or an equivalent fixative. The B-5 fixative contains mercuric chloride as well as formaldehyde, and it provides crisp nuclear detail. If a section is submitted in a mercury-based fixative, remember to notify your tissue processing laboratory personnel because these sections require special processing.

When submitting fresh tissue for special studies, collect the sample from solid ''fleshy'' areas of the tumor. Avoid areas that appear necrotic or sclerotic as these areas may not contain a sufficient quantity of viable tumor cells. The best techniques for submitting fresh tissue for immunophenotyping will depend on your individual laboratory, but in general a representative section of the node should be snap-frozen in optimal controlled temperature embedding medium for frozen tissue specimens (OCT) for immunohistochemical studies, and a separate 0.5- to 0.7-cm cube should be submitted fresh for flow cytometry. Again, the rapid handling of tissue for these studies is crucial, because delays can


Thinly slice
the lymph node.
1 mm³
Formalin
B5
Snap-frozen
fresh
Fresh
sterile
Glutaraldehyde
H&E, some
immunohisto-
chemistry,
chloracetate
esterase
H&E,
some
immunohisto-
chemistry
Full panel of
immunohisto-
chemistry,
enzyme
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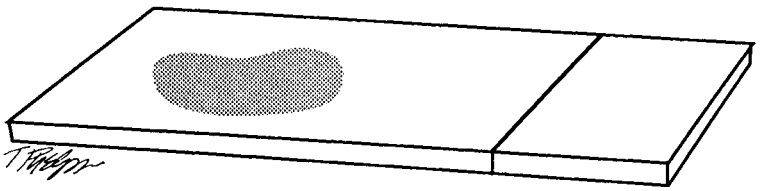

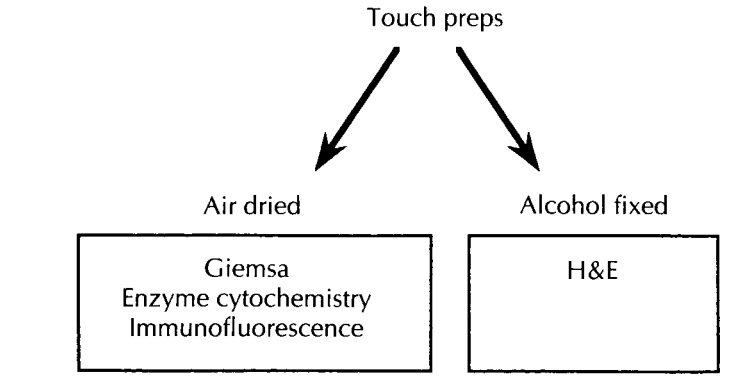
Touch preps
Air dried
Alcohol fixed
Giemsa
Enzyme cytochemistry
Immunofluorescence
H&E

result in diffusion artifacts during immunostaining. If tissue will be sent off-site for these analyses, it should not be frozen, but instead it should be kept cool on ice and rapidly transported.

If adequate tissue is available, and it usually is, fresh tissue should also be sent for genetic studies such as gene rearrangements and karyotyping. Submitting this tissue is important because antigen receptor gene rearrangement analysis may be required in those rare cases for which morphology and immunohistochemistry alone cannot establish the diagnosis. Obtain instructions on how to submit these specimens properly from your genetics laboratory.

Finally, if an infection is suspected or granulomas are encountered on a preliminary frozen section evaluation, fresh sterile tissue should be submitted for microbiologic studies. If a solid tumor is in the differential diagnosis, then consider placing a small piece of tissue into glutaraldehyde for electron microscopy.

Clearly, the take-home message here is that ''a stitch in time saves nine.'' Even the most challenging hematopoietic neoplasm can be diagnosed if well-fixed tissue for light microscopy and rapidly submitted fresh tissue for special studies are available. Table 41-1 summarizes the type of tissues to be submitted for specific staining methods and other analyses.

Important Issues to Address in Your Surgical Pathology Report on Lymph Nodes

- What procedure was performed, and what structures/organs are present?
- Anatomically, from where were the lymph nodes removed?
- What are the type and grade of the neoplasm?
- What are the number and size of the lymph nodes involved by tumor?
- What special studies were performed, and what were the results of these studies?

Extranodal Specimens

The lymphatic system is not limited to lymph nodes but encompasses diverse tissues and organs including the spleen, thymus, bone marrow, Waldeyer's ring, vermiform appendix, and mucosa-associated lymphoid tissue of the intestines and lung. Lymphomas can arise anywhere in this rather extensive lymphatic system. Moreover, they can arise in extranodal sites that are not part of the lymphatic system (e.g., thyroid, stomach). Although this chapter has focused on

TABLE 41-1. Tissues to submit.*

Tissue	Purpose
Air-dried touch imprints	Giemsa Enzyme cytochemistry—acid phosphatase, etc. Immunofluorescence for terminal transferase
Alcohol-fixed touch imprints	Hematoxylin and eosin
Formalin fixed, paraffin embedded	Hematoxylin and eosin Basic immunohistochemistry
B-5 fixed	Hematoxylin and eosin Basic immunohistochemistry
Snap-frozen fresh tissue	Detailed immunohistochemistry Enzyme histochemistry
Fresh sterile tissue	Cytogenetics Gene rearrangements Karyotyping Microbial cultures Flow cytometry
Glutaraldehyde	Electron microscopy

*Modified from Jaffee ES. *Surgical Pathology of the Lymph Nodes and Related Organs.* 2nd ed. Philadelphia, Pa: Saunders; 1995.

lymph nodes, it is important to recognize that a lymphoma can be encountered in almost any specimen.

If the nature of a tumor is unknown at the time of specimen processing, a touch prep or frozen section of the tumor is a fast, simple way to determine if you are dealing with lymphoid proliferation. This is important information to have as you begin the dissection because extranodal lymphoid proliferations, like their nodal counterparts, need to be submitted for special studies as appropriate. Once tissue has been obtained for special studies, the specimens can then be routinely processed in an organ-specific manner. There is no need to modify your approach in any significant way. Similar to dealing with some epithelial neoplasm, remember to document the dimensions of the tumor, determine the degree of involvement of adjacent structures, assess the status of the surgical margins, and evaluate the regional lymph nodes. The uninvolved tissues should also be sampled, and any additional pathologic processes (e.g., *Helicobacter pylori* infections in stomach resections, thyroiditis in thyroid resections) should be included in the final pathology report.