



Protocol for the Examination of Specimens From Patients With Hodgkin Lymphoma*

Version: Hodgkin 3.1.0.1

Protocol Posting Date: October 2013

This protocol is NOT required for accreditation purposes

*This protocol applies to Hodgkin lymphoma involving any site. The bone marrow protocol can also be used for reporting Hodgkin lymphoma involving the bone marrow.

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With guidance from the CAP Cancer and CAP Pathology Electronic Reporting Committees.

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Accreditation Requirements

This protocol can be utilized for clinical care purposes, but is not required for accreditation purposes.

CAP Laboratory Accreditation Program Protocol Required Use Date: Not applicable

Surgical Pathology Cancer Case Summary

Protocol posting date: October 2013

HODGKIN LYMPHOMA: Biopsy, Resection**Note: This case summary is recommended for reporting Hodgkin lymphoma specimens, but is not required for accreditation purposes.****Select a single response unless otherwise indicated.****Specimen (select all that apply) (Note A)**

- Lymph node(s)
 Other (specify): _____
 Not specified

Procedure

- Biopsy
 Resection
 Other (specify): _____
 Not specified

Tumor Site (select all that apply) (Note B)

- Lymph node(s), site not specified
 Lymph node(s)
 Specify site(s): _____
 Other tissue(s) or organ(s) (specify): _____
 Not specified

Histologic Type (based on the 2008 WHO classification) (Note C)

- Hodgkin lymphoma, histologic subtype cannot be determined
 Classical Hodgkin lymphoma, histologic subtype cannot be determined
 Nodular lymphocyte predominant Hodgkin lymphoma
 Nodular sclerosis classical Hodgkin lymphoma
 Mixed cellularity classical Hodgkin lymphoma
 Lymphocyte-rich classical Hodgkin lymphoma
 Lymphocyte-depleted classical Hodgkin lymphoma

+ Pathologic Extent of Tumor (select all that apply) (Note D)

- + Bone marrow involvement
 + Other site involvement
 + Specify site(s): _____

+ Additional Pathologic Findings

+ Specify: _____

Immunophenotyping (Immunohistochemistry) (Note E)

- Performed, see separate report: _____
 Performed
 Specify method(s) and results: _____
 Not performed

+ Clinical Prognostic Factors and Indices (select all that apply) (Note F)

- + ___ International Prognostic Score (IPS) (specify): _____
- + ___ B symptoms present
- + ___ Other (specify): _____

+ Comment(s)

Explanatory Notes

A. Specimen

Any number of specimen types may be submitted in the evaluation of Hodgkin lymphoma. Lymph nodes, mediastinal masses, bone marrow, spleen, lung, and liver are among the most common. Specimens submitted with a suspected diagnosis of Hodgkin lymphoma require special handling in order to optimize the diagnosis. Often, lymph node specimens are submitted where the differential diagnosis includes both Hodgkin and non-Hodgkin lymphomas, and, if possible, tissue should be obtained for possible molecular and other ancillary studies, which are often necessary for the diagnosis of non-Hodgkin lymphomas.^{1,2} Most flow cytometry, molecular, and cytogenetic studies will not aid in the diagnosis of Hodgkin lymphoma. Immunophenotyping by immunohistochemical staining is necessary in the initial diagnosis of nearly all cases of Hodgkin lymphoma. Because of this, well-fixed sections are of paramount importance. The guidelines detailed below are suggested for specimen handling in cases of suspected Hodgkin lymphoma.

- Tissue should be received fresh. Unsectioned lymph nodes should not be immersed in fixative, and care should be taken to make thin (2 mm) slices perpendicular to the long axis of the node to ensure optimal penetration of fixative.
- The fresh specimen size, color, and consistency should be recorded, as should the presence or absence of any visible nodularity, hemorrhage, or necrosis.
- Touch imprints may be made from the freshly cut surface, and the imprints fixed in alcohol or air dried. Unstained air-dried imprints can be used for fluorescence in situ hybridization (FISH) or other studies if necessary.
- For microbiology studies: submit a fresh portion of the lymph node (or other specimen type) sterilely in appropriate medium.
- Flow cytometry immunophenotyping is not routinely used in the diagnosis of Hodgkin lymphoma, but if the differential diagnosis includes non-Hodgkin lymphoma, a fresh portion of the specimen should be submitted in appropriate transport medium such as RPMI.
- Fixation (record fixative[s] used for individual slices of the specimen):
 - Estimated time from excision to fixation should be noted, if possible, as this may impact preservation or recovery of certain analytes such as RNA and phosphoproteins in fixed tissues.
 - Zinc formalin or B5 produces superior cytologic detail but is not suitable for DNA extraction and may impair some immunostains (eg, CD30). B5 also has the additional limitation of requiring proper hazardous materials disposal.
 - Formalin fixation is preferable when the tissue sample is limited, as it is most suitable for immunohistochemistry as well as many other ancillary tests such as molecular/genetic studies and in-situ hybridization.
 - Over-fixation (ie, more than 24 hours in formalin, more than 4 hours in zinc formalin or B5) should be avoided for optimal immunophenotypic reactivity.

B. Tumor Site

Hodgkin lymphomas are nearly always nodal based with cervical lymph nodes more commonly involved. It can also frequently be seen involving mediastinal, axillary, and paraaortic lymph nodes. Extranodal Hodgkin lymphoma can rarely be seen. The anatomic distribution of Hodgkin lymphoma, however, varies depending on the histologic type.³

C. Histologic Type

This protocol recommends assigning histologic type based on the World Health Organization (WHO) classification of lymphoid neoplasms.⁴ It was originally published in 2001 and more recently revised and updated in 2008.^{4,5} This classification encompasses both Hodgkin and non-Hodgkin lymphomas and allows distinction of individual lymphoid neoplasms based upon morphologic, immunophenotypic, cytogenetic, and clinical features. While histologic examination typically is thought to be the gold standard, the majority of Hodgkin lymphomas will require immunohistochemical staining, especially at the time of initial diagnoses.⁴⁻⁹ In addition, while Hodgkin lymphomas are currently divided into nodular lymphocyte predominant Hodgkin lymphoma and classical Hodgkin lymphomas (including nodular sclerosis, mixed cellularity, lymphocyte-rich, and lymphocyte-depleted subtypes), it should be recognized that classical Hodgkin lymphomas may not represent a single disease. In addition, there is overlap

between some cases of Hodgkin lymphoma and non-Hodgkin lymphoma, particularly diffuse large B-cell lymphomas (so-called gray zone lymphomas).^{4,10}

D. Pathologic Extent of Tumor (Stage)

The TNM classification is not used for staging Hodgkin lymphomas because the site of origin of the tumor is often unclear and there is no way to differentiate among T, N, and M. The Cotswold revision of the Ann Arbor staging classification is used for Hodgkin lymphoma.^{11,12} It was originally published over 30 years ago.

Historically, pathologic staging depended on the biopsy of multiple lymph nodes on both sides of the diaphragm, splenectomy, wedge liver biopsy, and bone marrow biopsy to assess distribution of disease. Currently, staging of Hodgkin lymphoma is more commonly clinical than pathologic. Clinical staging generally involves a combination of clinical, radiologic, and surgical data. Imaging studies (eg, computed tomography scans, magnetic resonance imaging studies, and positron emission tomography), biopsy (to determine diagnosis, histologic type, and extent of disease), and bone marrow examination are often the main criteria for staging Hodgkin lymphoma patients.

Cotswold Revision of the Ann Arbor Staging Classification of Hodgkin Lymphomas^{13,14}

Stage I	Involvement of a single lymph node region (I), or lymphoid structure (eg, spleen, thymus, Waldeyer's ring). [#]
Stage II	Involvement of 2 or more lymph node regions on the same side of the diaphragm (II) (the mediastinum is considered a single site). ^{##}
Stage III	Involvement of lymph node regions on both sides of the diaphragm (III) which may be accompanied by extralymphatic extension in association with lymph node involvement (IIIE) or splenic involvement (IIIS).
Stage IV	Involvement of extranodal site(s) beyond those designated E.

[#] Multifocal involvement of a single extralymphatic organ is classified as stage IE and not stage IV.

^{##} The number of lymph node regions involved may be indicated by a subscript: eg, II₃.

E designates involvement of a single extranodal site or contiguous or proximal known nodal site of disease.

E. Immunophenotyping

Immunophenotyping by flow cytometry and molecular testing by polymerase chain reaction (PCR) are currently not typically used or are not necessary for the diagnosis of Hodgkin lymphoma. Immunophenotyping using immunohistochemistry is necessary for the initial diagnosis of nearly all cases of Hodgkin lymphoma. It requires well-fixed tissue sections for optimal immunohistochemical staining and interpretation.

Immunophenotypes^{1,4-8}

The following is to be used as a guideline for the more common immunophenotype for each subtype of Hodgkin lymphoma. It is however, not entirely comprehensive and individual cases may vary somewhat in their immunophenotypic profile.

Nodular lymphocyte predominant Hodgkin lymphoma: Lymphocyte predominant cells (LP cells; previously called L&H cells) are CD20+, CD79a+, PAX5+, CD45+, BCL6+, OCT-2+, BOB.1+, EMA +/-, CD15-, CD30-, CD43-, EBER-.

Nodular sclerosis classical Hodgkin lymphoma: Classical Hodgkin/Reed-Sternberg cells are CD30+, CD15+/-, CD45-, PAX5+/-, CD20-/+ , CD79a-/+ , EBER-/+ , OCT-2-/+ , BOB.1-/+ , EMA-

Mixed cellularity classical Hodgkin lymphoma: Classical Hodgkin/Reed-Sternberg cells are CD30+, CD15+/-, CD45-, PAX5+/-, CD20-/+ , CD79a-/+ , EBER+/-, OCT-2-/+ , BOB.1-/+ , EMA-

Lymphocyte-rich classical Hodgkin lymphoma: Classical Hodgkin/Reed-Sternberg cells are CD30+, CD15+/-, CD45-, PAX5+/-, CD20-/+ , CD79a-/+ , EBER-/+ , OCT-2-/+ , BOB.1-/+ , EMA-

Lymphocyte-depleted classical Hodgkin lymphoma: Classical Hodgkin/Reed-Sternberg cells are CD30+, CD15+/-, CD45-, PAX5+/-, CD20-/+ , CD79a-/+ , EBER+/-, OCT-2-/+ , BOB.1-/+ , EMA-

F. Clinical Prognostic Factors and Indices

The International Prognostic Score (IPS) was developed for Hodgkin lymphoma to predict outcome based on the following adverse factors: serum albumin <4g/dL, hemoglobin concentration <10.5 g/dL, male sex, age ≥45 years, stage IV disease, white blood cell count ≥15,000/mm³, and lymphopenia <600/mm³ or <8%. The rate of freedom from progression by risk category is: 0 factors 84%, 1 factor 77%, 2 factors 67%, 3 factors 60%, 4 factors 51%, and 5 or more factors 42%.¹³

Although not always provided to the pathologist by the physician submitting the specimen, certain clinical findings are known to be of prognostic value in all stages of Hodgkin and non-Hodgkin lymphoma. In particular, systemic symptoms of fever (greater than 38°C), unexplained weight loss (more than 10% body weight) in the 6 months before diagnosis, and drenching night sweats are used to define 2 categories for each stage of lymphoma: A (symptoms absent) and B (symptoms present). The presence of B symptoms is known to correlate with extent of disease (stage and tumor bulk), but symptoms also have been shown to have prognostic significance for cause-specific survival that is independent of stage.¹³ In addition to the IPS, other prognostic factors, including HIV status, Bcl-2 expression, and pretreatment interleukin-10 serum levels, may be important.¹⁸⁻²¹

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