Protocol for the Examination of Specimens From Patients With Malignant Germ Cell and Sex Cord–Stromal Tumors of the Testis, Exclusive of Paratesticular Malignancies

A Basis for Checklists

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This protocol is intended to assist pathologists in providing clinically useful and relevant information as a result of the examination of surgical specimens. Use of this protocol is intended to be entirely voluntary. If equally valid protocols or similar documents are applicable, the pathologist is, of course, free to follow those authorities. Indeed, the ultimate judgment regarding the propriety of any specific procedure must be made by the physician in light of the individual circumstances presented by a specific patient or specimen.

It should be understood that adherence to this protocol will not guarantee a successful result. Nevertheless, pathologists are urged to familiarize themselves with this document. Should a physician choose to deviate from the protocol owing to the circumstances of a particular patient or specimen, the physician is advised to make a contemporaneous written notation of the reason for the procedure followed.

The College recognizes that this document may be used by hospitals, attorneys, managed care organizations, insurance carriers, and other payers. However, the document was developed solely as a tool to assist pathologists in the diagnostic process by providing information that reflects the state of relevant medical knowledge at the time the protocol was first published. It was not developed for credentialing, litigation, or reimbursement purposes. The College cautions that any uses of the protocol for these purposes involve considerations that are beyond the scope of this document.

PROTOCOL FOR THE EXAMINATION OF SPECIMENS FROM PATIENTS WITH MALIGNANT GERM CELL TUMORS AND SEX CORD–STROMAL TUMORS OF THE TESTIS, EXCLUSIVE OF PARATESTICULAR MALIGNANCIES

I. Biopsy

A. Clinical information

1. Patient identification
   a. Name
   b. Identification number
   c. Age (birth date)

2. Responsible physician(s)

3. Date of procedure

4. Other clinical information
   a. Relevant history
      (1) Previous cryptorchidism treated by orchiopexy
      (2) Previous contralateral testicular tumor treated by orchiectomy and lymphadenectomy
      (3) Other
   b. Relevant findings
      (1) Testicular enlargement or atrophy
      (2) Gynecomastia
      (3) Ambiguous genitalia, feminization, or other features of intersex syndrome
      (4) Serum levels of α-fetoprotein
      (5) Serum levels of β subunit of human chorionic gonadotropin (note A)
      (6) Imaging studies (eg, ultrasound, abdominal computerized tomograms, chest radiographs)
   c. Clinical diagnosis (or clinical indication/concern prompting biopsy)
   d. Procedure
      (1) Inguinal exposure with testicular isolation and biopsy
      (2) Transscrotal open or needle biopsy
      (3) Open or needle biopsy of metastatic site
   e. Operative findings
   f. Biopsy site(s)

B. Macroscopic examination

1. Specimen
   a. Unfixed/fixed (specify fixative)
   b. Size (3 dimensions)
   c. Number of pieces
   d. Orientation (if designated by surgeon)
   e. Descriptive features (eg, color/texture)
   f. Results of intraprocedural consultation

Accepted for publication August 26, 1998.
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This protocol was developed by the Cancer Committee of the College of American Pathologists and submitted for editorial review and publication. It represents the views of the Cancer Committee and is not the official policy of the College of American Pathologists.

Reprints: Joe Schramm, College of American Pathologists, 325 Waukegan Rd, Northfield, IL 60093-2750.
2. Submit all tissue for microscopic evaluation
3. Special studies (specify) (eg, electron microscopy, cytogenetics, molecular biologic studies, flow cytometry, image analysis)

C. Microscopic evaluation
1. Adequacy of specimen (if unsatisfactory for evaluation, specify reason)
2. Tumor
   a. Histologic type(s) (note B)
   b. Intratubular, invasive, or both
   c. Extent of invasion (note C)
      (1) Invasion or penetration of tunica albuginea (specify)
      (2) Involvement of paratesticular structures
   d. Blood/lymphatic vessel invasion (note D)
3. Other pathologic findings, if present (note E)
4. Results/status of special studies (specify)
5. Comments
   a. Correlation with intraprocedural consultation, as appropriate
   b. Correlation with other specimens, as appropriate
   c. Correlation with clinical information, as appropriate

II. Radical Orchiectomy
A. Clinical information
   1. Patient identification
      a. Name
      b. Identification number
      c. Age (birth date)
   2. Responsible physician(s)
   3. Date of procedure
   4. Other clinical information
      a. Relevant history
         (1) Previous cryptorchidism treated by orchiopexy
         (2) Previous contralateral testicular tumor treated by orchiectomy and lymphadenectomy
         (3) Other
      b. Relevant findings
         (1) Testicular enlargement or atrophy
         (2) Gynecomastia
         (3) Ambiguous genitalia, feminization, or other features of intersex syndrome
         (4) Serum levels of α-fetoprotein
         (5) Serum levels of β subunit of human chorionic gonadotropin (note A)
         (6) Imaging studies (eg, ultrasound, abdominal computerized tomograms, chest radiographs)
      c. Clinical diagnosis
      d. Procedure
      e. Operative findings
         (1) Laterality of testis
         (2) Inguinal or abdominal orchiectomy in cases of cryptorchidism

B. Macroscopic examination
   1. Specimen
      a. Organ(s)/tissue(s) included
      b. Unfixed/fixed (specify fixative)
      c. Dimensions, including length of spermatic cord

   d. External aspect
   e. Cut surface
   f. Results of intraoperative consultation
2. Tumor
   a. Location
   b. Size (3 dimensions)
   c. Descriptive characteristics (eg, color, hemorrhage, necrosis)
   d. Borders (circumscribed vs invasive)
   e. Extent of invasion (note C)
      (1) Description of intertunical fluid
      (2) Involvement of tunica vaginalis
      (3) Involvement of tunica albuginea
      (4) Involvement of spermatic cord
      (5) Involvement of paratesticular soft tissue
   f. Relation to resection margin(s), including spermatic cord
3. Additional pathologic findings, if present
   a. Tumor(s)
   b. Scars
   c. Calcification
   d. Other(s)
4. Tissues submitted for microscopic evaluation (note F)
5. Special studies (specify)

C. Microscopic evaluation
   1. Tumor
      a. Histologic type (estimate percentage of each component for mixed tumors) (note B)
      b. Intratubular, invasive, or both
      c. Extent of invasion (note C)
         (1) Invasion or penetration of tunica albuginea (specify)
         (2) Involvement of paratesticular structures
      d. Lymphatic/blood vessel invasion (specify if in testis or paratestis/spermatic cord)
   2. Status of resection margin(s), including spermatic cord
   3. Additional pathologic findings, if present (note E)
   4. Regional lymph nodes
      a. Number present
      b. Number involved by tumor
   5. Other tissue(s)
      a. Involved by tumor
      b. Uninvolved by tumor
   6. Results/status of special studies (specify)
7. Comments
   a. Correlation with intraoperative consultation, as appropriate
   b. Correlation with other specimens, as appropriate
   c. Correlation with clinical information, as appropriate

III. Retroperitoneal Lymphadenectomy
A. Clinical information
   1. Patient identification
      a. Name
      b. Identification number
      c. Age (birth date)
   2. Responsible physician(s)
   3. Date of procedure
   4. Regional lymph nodes
      a. Number present
      b. Number involved by tumor
   5. Other tissue(s)
      a. Involved by tumor
      b. Uninvolved by tumor
   6. Results/status of special studies (specify)
7. Comments
   a. Correlation with intraoperative consultation, as appropriate
   b. Correlation with other specimens, as appropriate
   c. Correlation with clinical information, as appropriate
4. Other clinical information
   a. Relevant history
      (1) Previous cryptorchidism treated by orchiopexy
      (2) Previous contralateral testicular tumor treated by orchiectomy and lymphadenectomy
      (3) Other
   b. Relevant findings
      (1) Testicular enlargement or atrophy
      (2) Gynecomastia
      (3) Ambiguous genitalia, feminization, or other features of intersex syndrome
      (4) Serum levels of \( \alpha \)-fetoprotein
      (5) Serum levels of \( \beta \) subunit of human chorionic gonadotropin (note A)
      (6) Imaging studies (eg, ultrasound, abdominal computerized tomograms, chest radiographs)
   c. Clinical diagnosis
   d. Procedure (eg, radical, nerve-sparing, or other form of retroperitoneal lymphadenectomy)
   e. Operative findings
   f. Anatomic site(s) of specimen(s)

B. Macroscopic examination
   1. Specimen
      a. Organ(s)/tissues included
      b. Unfixed/fixed (specify fixative)
      c. Results of intraoperative consultation
   2. Regional lymph nodes
      a. Number of lymph node groups and site of each
      b. For each nodal group
         (1) Size (3 dimensions) of nodal group
         (2) Number of lymph nodes identified
         (3) Number of lymph nodes involved by tumor
            i. Size ranges of identifiable tumor nodules or dimensions of tumor-matted nodes
            ii. Descriptive features of tumor, if present (eg, color, hemorrhage, necrosis)
   3. Spermatic cord structures, if present
      a. Descriptive characteristics
      b. Involvement by tumor
   4. Tissues submitted for microscopic evaluation (note F)
      a. All nodal groups
         (1) Number of lymph nodes identified per group
         (2) Number of lymph nodes submitted for each group
      b. Spermatic cord structures
      c. Frozen section tissue fragment(s) (unless saved for special studies)
   5. Special studies (specify)

C. Microscopic evaluation
   1. Regional lymph nodes
      a. Number of lymph nodes in each nodal group
      b. Number involved by tumor in each nodal group
         (1) Histologic type(s) (note B)
         (2) Extent of nodal replacement
            i. Microfocus
            ii. Multiple microfoci
            iii. Scattered nodules
            iv. Diffuse replacement
         (3) Involvement of extranodal soft tissues, including residual spermatic cord
         (4) Necrosis, if present
         (5) Associated scar tissue
   2. Results/status of special studies (specify)
   3. Comments
      a. Correlation with intraoperative consultation, as appropriate
      b. Correlation with other specimens, as appropriate
      c. Correlation with clinical information, as appropriate

IV. Cytologic Evaluation of Metastatic Sites (note G)
A. Clinical information
   1. Patient identification
      a. Name
      b. Identification number
      c. Age (birth date)
   2. Responsible physician
   3. Date of procedure
   4. Clinical information
      a. Relevant history
         (1) Previous diagnosis and treatment
         (2) Histopathologic type(s) of prior testicular tumor
      b. Relevant findings
         (1) Clinical findings
         (2) Marker studies (eg, \( \alpha \)-fetoprotein, \( \beta \)-human chorionic gonadotropin)
         (3) Imaging studies (solid or cystic lesion)
   5. Clinical diagnosis
   6. Procedure (eg, fine-needle aspiration)
   7. Anatomic site(s) of specimen(s)

B. Macroscopic examination
   1. Specimen
      a. Unfixed/fixed (specify fixative)
      b. Number and appearance of slides prepared/received
         (1) Air dried and/or wet fixed
         (2) Grossly bloody
         (3) Tissue fragments
      c. Quantity and appearance of fluid, if appropriate
      d. Results of intraprocedural consultation
   2. Material submitted for microscopic evaluation (eg, smear, cytocentrifuge, touch or filter preparations, cell block)
   3. Special studies (specify)

C. Microscopic evaluation
   1. Adequacy of specimen (if unsatisfactory for evaluation, specify reason)
   2. Tumor, if present (specify types, if possible) (note H)
   3. Other pathologic findings
      a. Inflammation
      b. Necrosis
      c. Fibrosis
      d. Foam cell reaction
      e. Other
   4. Results/status of special studies (specify)
5. Comments
   a. Correlation with intraoperative consultation, as appropriate
   b. Correlation with clinical information, as appropriate
   c. Correlation with other specimens, as appropriate

EXPLANATORY NOTES

A: Serum Markers.—The protocol emphasizes the importance of relevant clinical information in the pathologic evaluation of specimens. Serum marker studies play a key role in the clinical management of patients with testicular germ cell tumors. For the occurrence of elevated serum levels of α-fetoprotein or the β subunit of human chorionic gonadotropin may indicate the need for additional sections of certain specimens if the initial findings do not account for such elevations. As another example, gynecomastia accompanies tumors in the sex cord–stromal category more frequently than germ cell tumors, and an awareness of this clinical finding alerts the pathologist to consider the possibility of a sex cord–stromal tumor.

B: Histologic Type.—The protocol applies to malignant tumors of the testis, the vast majority of which are of germ cell origin. It may also be applied to other malignant or potentially malignant tumors of the testis included in the following World Health Organization classification. For lymphomas and plasmacytomas of the testis, refer to the non-Hodgkin’s lymphoma protocol.

World Health Organization Histologic Classification of Testicular Tumors

Germ cell tumors

Precursor lesion

Intratubular germ cell neoplasm, unclassified

Tumors of one histologic type

Seminoma

Variant: seminoma with syncytiotrophoblastic cells

Spermatocytic seminoma

Variant: spermatocytic seminoma with a sarcomatous component

Embryonal carcinoma

Yolk sac tumor

Choriocarcinoma

Variant: “monophasic” type

Placental site trophoblastic tumor

Teratoma

Matue

Immature

With a secondary malignant component

Monodermal variants

Carcinoid

Primitive neuroectodermal tumor

Tumors of more than 1 histologic type

Mixed germ cell tumor (specify components; estimate percentage)

Polyembryoma

Diffuse embryoma

Sex cord–stromal tumors

Leydig cell tumor

Sertoli cell tumor

Variant: large cell calcifying Sertoli cell tumor

Variant: sclerosing Sertoli cell tumor

Granulosa cell tumor

Adult type

Juvenile type

Mixed and indeterminate (unclassified) sex cord–stromal tumor

Mixed germ cell–sex cord–stromal tumors

Gonadoblastoma

Others

Miscellaneous

Sarcoma (specify type)

Lymphoma (specify type)

Granulocytic sarcoma or leukemic infiltrates

Adenocarcinoma of rete testis

Carcinomas and borderline tumors of ovarian type

Malignant mesothelioma

C: Staging.—The protocol recommends staging according to the American Joint Committee on Cancer (AJCC) and the International Union Against Cancer (UICC) TNM Staging System. Additional criteria for staging seminomas according to a modification of the Royal Marsden system are also recommended. Some studies suggest that the staging of disease in patients with seminoma by the TNM system is less meaningful therapeutically than staging by a modification of the Royal Marsden method. The latter staging system subdivides cases with retroperitoneal metastases into several groups according to the total tumor dimension rather than the size of the largest lymph node, as in the TNM system. Also, the data from a large Danish study of seminomas clinically limited to the testis do not support the conclusion that local staging of the primary tumor, as performed in the TNM system, provides useful prognostic information; rather, the most valuable prognostic indicator was the size of the seminoma. This protocol therefore encourages the use of both the TNM system and the modified Royal Marsden staging system for patients with seminoma.

AJCC/UICC TNM and Stage Groupings

Primary Tumor (T)*

TX Primary tumor cannot be assessed

T0 No evidence of primary tumor (eg, histologic scar in testis)

Tis Intratubular germ cell neoplasia (“carcinoma” in situ)

T1 Tumor limited to the testis and epididymis without vascular/lymphatic invasion (tumor may invade tunica albuginea but not tunica vaginalis)

T2 Tumor limited to the testis and epididymis with vascular/lymphatic invasion or tumor extending through tunica albuginea with involvement of tunica vaginalis

T3 Tumor invades spermatic cord with or without vascular/lymphatic invasion

T4 Tumor invades scrotum with or without vascular/lymphatic invasion

* By AJCC/UICC convention, the designation “T” of the TNM classification refers exclusively to the first resection of a primary tumor. The prefix symbol “p” refers to the pathologic classification of the TNM (pTNM), as opposed to the clinical classification. Pathologic classification is based on gross and microscopic examination. Therefore, pT entails a resection of the primary tumor or biopsy adequate to evaluate the highest pT category; pN entails removal of nodes adequate to validate lymph node metas-
tasis; and pM implies microscopic examination of distant lesions. Clinical classification (cTNM) is usually carried out by the referring physician before treatment during initial evaluation of the patient or when pathologic classification is not possible.

The absence or presence of residual tumor following preoperative, nonsurgical therapy (eg, chemotherapy or radiation treatment) may be described by the symbol “R” (eg, rpT1). Local recurrence following a previous resection by the TNM classification preceded by the symbol “y” (eg, ypT1). If residual tumor is present, its extent may be documented and is classified as follows:

- RX: Presence of residual tumor cannot be assessed
- R0: No residual tumor
- R1: Microscopic residual tumor
- R2: Macroscopic residual tumor

If residual tumor is present, its extent may be documented by the TNM classification preceded by the symbol “y” (eg, ypT1). Local recurrence following a previous resection should be classified with the prefix “r” (eg, rpT1).

**Regional Lymph Nodes (N)**

- NX: Regional nodes cannot be assessed
- N0: No regional lymph node metastasis
- N1: Metastasis with a lymph node mass ≤2 cm in greatest dimension and ≤5 positive nodes, none >2 cm in greatest dimension
- N2: Metastasis with a lymph node mass >2 cm but ≤5 cm in greatest dimension, or >5 positive nodes, none >5 cm, or evidence of extranodal extension of tumor
- N3: Metastasis with a lymph node mass >5 cm in greatest dimension

**Distant Metastasis (M)**

- MX: Presence of distant metastasis cannot be assessed
- M0: No distant metastasis
- M1: Distant metastasis present
  - M1a: Nonregional lymph node or pulmonary metastasis
  - M1b: Distant metastasis other than to nonregional lymph nodes and lungs

**Stage Groupings**

- Stage 0: pTis
- Stage I: pT1–4
- Stage II: Any pT/TX

**Serum Tumor Markers (S)**

- SX: Serum marker studies not available or performed
- S0: Serum marker study levels within normal limits

**Human Gonadotropin, mL/U/mL**

<table>
<thead>
<tr>
<th>Stage</th>
<th>Lactate Dehydrogenase</th>
<th>α-Fetoprotein, ng/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td>S1</td>
<td>&lt;1.5 × N+ and</td>
<td>&lt;5000 and &lt;1000</td>
</tr>
<tr>
<td>S2</td>
<td>1.5–10 × N+ or</td>
<td>5000–50000 or 1000–10000</td>
</tr>
<tr>
<td>S3</td>
<td>&gt;10 × N+ or</td>
<td>&gt;50000 or &gt;100000</td>
</tr>
</tbody>
</table>

† N indicates the upper limit of normal for the lactate dehydrogenase assay.

**Stage Groupings**

- Stage 0: pTis
- Stage I: pT1–4
- Stage II: Any pT/TX

**Modified Royal Marsden Staging System**

<table>
<thead>
<tr>
<th>Stage</th>
<th>Tumor confined to the testis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stage I</td>
<td>Tumor confined to the testis</td>
</tr>
<tr>
<td>Stage II</td>
<td>Infradiaphragmatic nodal involvement</td>
</tr>
<tr>
<td>Stage III</td>
<td>Any pT/TX Any N M1, M1a SX</td>
</tr>
<tr>
<td>Stage IV</td>
<td>Extraneal metastasis</td>
</tr>
</tbody>
</table>

**D: Blood/Lymphatic Vessel Invasion**

- D1: Blood/Lymphatic vessel invasion

**E: Additional Pathologic Findings**

- E1: Additional Pathologic Findings

**F: Tissues Submitted for Microscopic Evaluation**

The entire testicular tumor may be blocked if it requires 10 blocks or fewer (tissue may be retained for special studies); 10 blocks of larger tumors may be taken unless the tumor is >10 cm, in which case 1 block may be submitted for every 1 cm of maximum tumor dimension. Some blocks should contain the interface with nontumorous testis because lymphatic invasion is best appreciated there. Tissues to be sampled include all of the grossly different.
types of tumor; testicular hilus; uninvolved testis; spermatic cord, including cord margin; epididymis; other lesion(s); all identifiable lymph nodes*; and other tissue(s) submitted with the specimen.

*For large masses that have obliterated individual nodes, 1 section for every centimeter of maximum tumor dimension, especially fleshy-appearing foci, may be taken.

G: Cytology.—The role of cytologic diagnosis in patients with testicular tumors is ordinarily limited to the investigation of possible metastatic lesions, usually by fine-needle aspiration. The cytologic investigation by fine-needle aspiration of the primary testicular neoplasm is unnecessary because orchietomy must be performed for proper classification and treatment. Furthermore, the cytologic diagnosis is often incomplete owing to sampling limitations and the frequently heterogeneous nature of testicular tumors.

H: Metastatic Teratoma.—Often the most important distinction in patients with metastatic testicular germ cell tumor, particularly following initial chemotherapy, is the differentiation of metastatic teratoma from nonteratomatous types of germ cell tumor. Pure teratomatous metastasis is generally treated by surgical excision, whereas patients who have metastatic embryonal carcinoma, yolk sac tumor, etc, are usually treated with chemotherapy. For cytologic diagnosis, specify seminoma versus nonseminomatous germ cell tumor, and teratomatous versus nonteratomatous germ cell tumor, if possible. Illustrative diagnoses are as follows: (1) positive for malignant cells from a nonseminomatous germ cell tumor, consistent with metastatic embryonal carcinoma; (2) positive for malignant cells from a nonseminomatous germ cell tumor, consistent with metastatic teratoma; (3) positive for malignant cells from a nonseminomatous germ cell tumor, further classification not possible; and (4) positive for malignant cells, consistent with metastatic germ cell tumor, further classification not possible.

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References

Bibliography