Protocol for the Examination of Biopsy Specimens From Patients With Gastrointestinal Stromal Tumor (GIST)

Version: 4.2.0.0
Protocol Posting Date: June 2021
The use of this protocol is recommended for clinical care purposes but is not required for accreditation purposes.

This protocol may be used for the following procedures AND tumor types:

<table>
<thead>
<tr>
<th>Procedure</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Biopsy</td>
<td></td>
</tr>
<tr>
<td>Tumor Type</td>
<td>Description</td>
</tr>
<tr>
<td>Gastrointestinal stromal tumor</td>
<td></td>
</tr>
</tbody>
</table>

The following should NOT be reported using this protocol:

<table>
<thead>
<tr>
<th>Procedure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Resection</td>
</tr>
<tr>
<td>Cytologic specimens</td>
</tr>
</tbody>
</table>

Authors
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With guidance from the CAP Cancer and CAP Pathology Electronic Reporting Committees.
* Denotes primary author.
Accreditation Requirements
The use of this case summary is recommended for clinical care purposes but is not required for accreditation purposes. The core and conditional data elements are routinely reported. Non-core data elements are indicated with a plus sign (+) to allow for reporting information that may be of clinical value.

Summary of Changes

v 4.2.0.0

- General Reformatting
Reporting Template

Protocol Posting Date: June 2021
Select a single response unless otherwise indicated.

CASE SUMMARY: (GASTROINTESTINAL STROMAL TUMOR (GIST): Biopsy)
Standard(s): AJCC-UICC 8
This case summary is recommended for reporting biopsy specimens, but is not required for accreditation purposes.

CLINICAL

+Prebiopsy Treatment (select all that apply)
___ No known prebiopsy therapy
___ Systemic therapy performed (specify type): _________________
___ Therapy performed, type not specified
___ Not specified

SPECIMEN

Procedure
___ Core needle biopsy
___ Endoscopic biopsy
___ Other (specify): _________________
___ Not specified

TUMOR

Tumor Site (Note A)
___ Esophagus: _________________
___ Cervical esophagus
___ Thoracic esophagus
___ Abdominal esophagus
___ Upper third of esophagus
___ Middle third of esophagus
___ Lower third of esophagus
___ Esophagus, NOS
___ Stomach: _________________
___ Cardia of stomach
___ Fundus of stomach
___ Body of stomach
___ Gastric antrum
___ Pylorus
___ Lesser curvature of stomach, NOS
___ Greater curvature of stomach, NOS
___ Stomach, NOS
___ Small Intestine: _________________
___ Duodenum
___ Jejunum
___ Ileum (excluding ileocecal valve)
___ Meckel diverticulum (site of neoplasm)
___ Small intestine, NOS
___ Appendix: ____________________
___ Colon: ______________________
___ Cecum
___ Ascending colon
___ Hepatic flexure of colon
___ Transverse colon
___ Splenic flexure of colon
___ Descending colon
___ Sigmoid colon
___ Colon, NOS
___ Rectosigmoid junction: _________________
___ Rectum, NOS: _________________
___ Retroperitoneum: _________________
___ Peritoneum, including omentum and mesentery (specify parts): _________________
___ Peritoneum, NOS: _________________
___ Other (specify): ____________________
___ Cannot be determined: _________________
___ Not specified

Histologic Type
___ Gastrointestinal stromal tumor, spindle cell type
___ Gastrointestinal stromal tumor, epithelioid type
___ Gastrointestinal stromal tumor, mixed
___ Gastrointestinal stromal tumor, other (specify): _________________
+Histologic Type Comment: ____________________

Mitotic Rate (Note B)
The required total count of mitoses is per 5 mm$^2$ on the glass slide section. With the use of older model microscopes, 50 HPF is equivalent to 5 mm$^2$. Most modern microscopes with wider 40X lenses / fields require approximately 20 to 25 HPF to encompass 5 mm$^2$. If necessary please measure field of view to accurately determine actual number of fields required to be counted on individual microscopes to encompass 5 mm$^2$.
___ Specify mitotic rate per 5 mm$^2$: _________________ mitoses per 5 mm$^2$
___ Other (specify): ____________________
___ Cannot be determined (explain): ____________________

Histologic Grade (Note B)
___ G1, low grade (mitotic rate less than or equal to 5 per 5 mm$^2$)
___ G2, high grade (mitotic rate greater than 5 per 5 mm$^2$)
___ Other (specify): _________________
___ GX, cannot be assessed: _________________

+Necrosis
___ Not identified
___ Present
+Extent of Necrosis
___ Specify percentage: _________________ %
___ Other (specify): _________________
___ Cannot be determined: _________________
___ Cannot be determined: _________________
Treatment Effect (Note C)
___ No known prebiopsy therapy
___ Not identified
___ Present

+Percentage of Viable Tumor
___ Specify percentage: _________________ %
___ Other (specify): _________________
___ Cannot be determined: _________________
___ Cannot be determined: _________________

Risk Assessment (Note D)
___ None
___ Very low risk
___ Low risk
___ Moderate risk
___ High risk
___ Overtly metastatic
___ Cannot be determined: _________________

+Tumor Comment: _________________

ADDITIONAL FINDINGS

+Additional Findings (specify): _________________

SPECIAL STUDIES (Note E)
The CAP GIST Biomarker Template can be used for reporting biomarkers. Pending biomarker studies should be listed in the Comments section of this report.

+Immunohistochemical Studies (select all that apply)
___ Not performed
___ KIT (CD117)
   KIT (CD117)
___ Positive
___ Negative
___ Pending
___ DOG1 (ANO1)
   DOG1 (ANO1)
___ Positive
___ Negative
___ Pending
___ SDHB
   SDHB
___ Intact
___ Deficient
___ Pending
___ SDHA
   SDHA
___ Intact
___ Deficient
___ Pending
___ Other (specify): ____________________

+Molecular Genetic Studies (e.g., KIT, PDGFRA, BRAF, SDHA/B/C/D, or NF1 mutational analysis)
___ Submitted for analysis; results pending
___ Performed, see separate report: _________________
___ Performed (specify method(s) and result(s)): _________________
___ Not performed

COMMENTS

Comment(s): ____________________
Explanatory Notes

A. Location
Gastrointestinal stromal tumors may occur anywhere along the entire length of the tubal gut, as well as in extravisceral locations, which include the omentum, mesentery, pelvis, and retroperitoneum. Typically, they arise from the wall of the gut and extend inward toward the mucosa, outward toward the serosa, or in both directions. Lesions that involve the wall of the gastrointestinal (GI) tract frequently cause ulceration of the overlying mucosa. Infrequently, lesions invade through the muscularis mucosae to involve the mucosae. Mucosal invasion is an adverse prognostic factor in numerous studies. Because the anatomic location along the GI tract affects prognosis, with location in the stomach having a more favorable prognosis, it is very important to specify anatomic location as precisely as possible.  

References

B. Histologic Grade
Histologic grading, an important component of soft tissue sarcoma staging, is not well suited to GISTs, because most of these tumors have low or relatively low mitotic rates below the thresholds used for grading of soft tissue tumors, and because GISTs often manifest aggressive features with mitotic rates below the thresholds used for soft tissue tumor grading (the lowest tier of mitotic rates for soft tissue sarcomas being 10 mitoses per 10 HPF). In GIST staging, the grade is determined entirely by mitotic activity.  

GX: Grade cannot be assessed
G1: Low grade; mitotic rate ≤5/5 mm²
G2: High grade; mitotic rate >5/5 mm²

The mitotic count should be initiated on an area that on screening magnification shows the highest level of mitotic activity and be performed as consecutive high-power fields (HPF). Stringent criteria should be applied when counting mitotic figures; pyknotic, dyskaryotic or apoptotic nuclei should not be regarded as mitosis.

Note: The required total count of mitoses is per 5 mm² on the glass slide section. With the use of older model microscopes, 50 HPF is equivalent to 5 mm². Most modern microscopes with wider 40X lenses/fields require approximately 20 to 25 HPF to encompass 5 mm². If necessary, please measure
field of view to accurately determine actual number of fields required to be counted on individual microscopes to encompass 5 mm².

References

C. Treatment Effect
Gastrointestinal stromal tumors respond well to the newer targeted systemic therapies, imatinib mesylate and sunitib malate. The types of treatment effects that have been seen are hypocellularity, myxoid stroma, fibrosis, and necrosis. Nests of viable tumor cells are virtually always seen. Because all of these histologic features can be seen in untreated GISTs, it is not possible to know whether they are due to treatment or not. As a practical compromise, we think it is best to report the percentage of viable tumor after treatment.

D. Risk Assessment
Because GISTs can recur many years after initial excision, we now regard most GISTs as having at least some potential for distant metastasis. This concept was originally the result of a National Cancer Institute-sponsored consensus conference that was held in 2002.1 More specific data generated by large follow-up studies refined the biologic potential assessment2,3,4,5,6 Criteria obtained from those data were adopted in a National Cancer Care Network (NCCN) Task Force report on GIST.7 We have adopted the criteria for risk stratification, as indicated in Table 1.2,3,4,5,6 The scheme includes anatomic site as a factor, because small bowel GISTs carry a higher risk of progression than gastric GISTs of similar size and mitotic activity. This prognostic assessment applies best to KIT/PDGFRA mutant GISTs whereas SDD-deficient GISTS are more unpredictable.8 For anatomic sites not listed in this table, such as esophagus, mesentery, and peritoneum, or in the case of “insufficient data,” it is best to use risk criteria for jejunum/ileum.

Table 1. Guidelines for Risk Assessment of Primary Gastrointestinal Stromal Tumor (GIST)

<table>
<thead>
<tr>
<th>Tumor Parameters</th>
<th>Risk of Progressive Disease (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Gastric</td>
</tr>
<tr>
<td>Mitotic Rate</td>
<td>≤2 cm</td>
</tr>
<tr>
<td></td>
<td>&gt;2 - ≤5 cm</td>
</tr>
<tr>
<td></td>
<td>&gt;5 - ≤10 cm</td>
</tr>
<tr>
<td></td>
<td>&gt;10 cm</td>
</tr>
<tr>
<td>Size</td>
<td>≤2 cm</td>
</tr>
<tr>
<td></td>
<td>&gt;2 - ≤5 cm</td>
</tr>
<tr>
<td></td>
<td>&gt;5 - ≤10 cm</td>
</tr>
<tr>
<td></td>
<td>&gt;10 cm</td>
</tr>
</tbody>
</table>
Adapted with permission from Miettinen and Lasota. Copyright 2006 by Elsevier.

Defined as metastasis or tumor-related death.

Denotes small number of cases.

Data based on long-term follow-up of 1055 gastric, 629 small intestinal, 144 duodenal, and 111 rectal GISTs from the pre-imatinib era.

Note: See Note B, “Histologic Grade,” regarding the number of high power fields to evaluate.

References

E. Ancillary Studies

Immunohistochemistry
Because of the advent of small-molecule kinase inhibitor therapy in the treatment of GIST (see the following), it has become imperative to distinguish GIST from its histologic mimics, mainly leiomyoma, leiomyosarcoma, schwannoma, and desmoid fibromatosis. Immunohistochemistry is instrumental in the workup of GIST. For the initial work up of GIST, a basic immunohistochemical panel including CD117 (KIT), DOG1 (Ano1), Desmin, S100 protein and CD34 is recommended. GISTs are immunoreactive for KIT (CD117) (approximately 95%) and/or DOG1(>99%). KIT immunoreactivity is usually strong and diffuse but can be more focal in unusual cases (Figure 1, A and B). It is not unusual for GISTs to exhibit dot-like perinuclear staining (Figure 1, C), while less commonly, some cases exhibit membranous staining (Figure 1, D). These patterns do not clearly correlate with mutation type or response to therapy. Most KIT-negative / DOG1 positive GISTs are gastric or extra-visceral GISTs and almost invariably harbor a platelet-derived growth factor receptor A (PDGFRα) mutation. DOG1 expression is not related to mutational status in GISTs, and it may be a useful marker to identify a subset of patients with CD117-negative GISTs, who might benefit from targeted therapy. Approximately 70% of GISTs are positive for CD34, 30% to 40% are positive for smooth muscle actin, 5% are positive for S100 protein (usually focal), 5% are positive for desmin (usually focal), and 1% to 2% are positive for keratin (weak/focal).
Since succinate dehydrogenase (SDH)-deficient GISTs have specific implications (see the following), it is recommended to screen all gastric GISTs for loss of SDH by immunohistochemistry, usually best accomplished by staining for SDHB, which is loss in all subtypes of SDH-deficient GISTs.\textsuperscript{8,9,10,11} Mutations in SDHA are detected in 30\% of SDH-deficient GISTs and loss of expression of SDHA specifically identifies tumors with SDHA mutations; other SDH-deficient GISTs show normal (intact) cytoplasmic staining for SDHA.\textsuperscript{12,13} Patients with SDH-deficient GIST should be referred to a genetic counselor for appropriate work up.

**Figure 1.** Patterns of KIT staining in gastrointestinal stromal tumor (GIST). A. Diffuse and strong immunoreactivity in a typical GIST. B. Focal and weak pattern in an epithelioid gastric GIST with a PDGFRA mutation. C. Dot-like perinuclear staining. D. Membranous pattern. (Original magnification X400.)

**Molecular Analysis**
Approximately 75\% of GISTs possess activating mutations in the KIT gene, whereas another 10\% have activating mutations in the PDGFRA gene.\textsuperscript{14,15,16,17} These mutations result in virtually full-length KIT proteins that exhibit ligand-independent activation. KIT and PDGFRA each contain 21 exons. However, mutations cluster within "hotspots": exons 9, 11, 13, and 17 in KIT, and exons 12, 14, and 18 in PDGFRA (Figure 2). About 5\% to 10\% of GISTs appear to be negative for both KIT and PDGFRA mutations. The most recent NCCN Task Force on GIST strongly encourages that KIT and PDGFRA mutational analysis be performed if tyrosine kinase inhibitors (TKIs) are considered as part of the treatment plan for unresectable or metastatic disease and that mutational analysis be considered for patients with primary disease, particularly those with high-risk tumors. KIT and PDGFRA mutation status can be determined easily from paraffin-embedded tissue. Secondary or acquired mutations can be associated with development of tumor resistance in the setting of long-term imatinib mesylate treatment. These are usually point mutations that occur most commonly in KIT exons 13, 14, and 17.\textsuperscript{18} The clinical utility of these mutations is an evolving concept, but it is important not to confuse them with the primary or initial mutation in GIST.

Recent studies focusing on the molecular classification of GISTs recognized two major subgroups: succinate dehydrogenase (SHD)-competent and SDH-deficient GISTs, both of which can arise in the sporadic or familiar setting.\textsuperscript{8,9} SDH-competent GISTs include tumors with mutations of KIT and PDGFRA...
as well of a subset of wild-type GISTs with mutations mainly in NF1 and BRAF genes. On the other hand, 
SDH-deficient GISTs include tumors with a genetic alteration in any of the SDH subunits leading to SDH 
dysfunction.

SDH-deficient GISTs represent approximately 8% of GISTs and comprise some sporadic cases, the 
majority of pediatric GISTs, and two forms of syndromic GISTs (Carney triad and Carney-Stratakis 
syndrome). SDH is a mitochondrial enzyme comprising four subunits (SDHA, SDHB, SDHC and SDHD) 
that is involved in the Krebs cycle. Genetic alteration of any of the four subunits results in SDH 
dysfunction and subsequent loss of SDHB expression by immunohistochemistry. SDH deficient GISTs 
arise almost exclusive in the stomach, affect predominantly female patients and tend to manifest at a 
young age. Pathologic features associated with SDH-deficient tumors include multinodular and/or 
plexiform growth pattern, epithelioid morphology, lymphovascular invasion, nodal involvement and 
frequent metastasis to the liver and peritoneum. Importantly, germline mutations in the genes coding for 
any of the SHD subunits can lead to paragangliomas/pheochromocytomas, SDH-deficient renal cell 
carcinoma and pituitary tumors in addition to GISTs. Since SDH-deficient GISTs typically require germline 
genetic testing possibly including family members as well as possible surveillance for 
paragangliomas/pheochromocytomas, it is recommended that all gastric GISTs be screened for loss of 
SDHB by immunohistochemistry. All patients with SDH-deficient GISTs identified by loss of SDHB stain 
should be referred to a genetic counselor.

Figure 2. Locations and frequency of activating KIT and PDGFRA mutations in GIST. Adapted with 
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reserved.

KIT and PDGFRA are excellent targets for small-molecule tyrosine kinase inhibitors, and two compounds 
of this class, imatinib mesylate (Gleevec, Novartis Pharmaceuticals, Basel, Switzerland) and sunitinib 
malate (Sutent, Pfizer Pharmaceuticals, New York, New York), have shown efficacy in clinical trials and 
have been approved by the US Food and Drug Administration for the treatment of GIST. [19,20,21] SDH-
deficient GISTs are usually resistant to imatinib but may have a higher probability of response to 
sunitinib. [8] Because different tyrosine kinase inhibitors (TKIs) may have more efficacy in genetic subsets 
of GIST, oncologists may want to know the mutation status of each GIST, because this may impact which 
drug each patient should receive. [14,22] Secondary resistance mutations may also affect drug selection as 
their significance is further defined.
References


