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

Version: GIST Biopsy 4.1.0.0 **Protocol Posting Date:** August 2019

Accreditation Requirements
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:

Procedure	Description
Biopsy	
Tumor Type	**Description**
Gastrointestinal stromal tumor	

The following should NOT be reported using this protocol:

Procedure
Resection
Cytologic specimens

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Summary of Changes

Version 4.1.0.0
Resection and biopsy case summaries separated into discrete cancer protocols

The following were modified:
Ancillary Testing - included SDHB and SDHA
Treatment Effect - No known prebiopsy therapy

Surgical Pathology Cancer Case Summary

Protocol posting date: August 2019

GASTROINTESTINAL STROMAL TUMOR (GIST): Biopsy

Note: This case summary is recommended for reporting biopsy specimens, but is not required for accreditation purposes. Core data elements are bolded to help identify routinely reported elements.

Select a single response unless otherwise indicated.

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

Tumor Site (Note A)
Specify: ______________________________
___ Not specified

Histologic Type
___ Gastrointestinal stromal tumor, spindle cell type
___ Gastrointestinal stromal tumor, epithelioid type
___ Gastrointestinal stromal tumor, mixed
___ Gastrointestinal stromal tumor, other (specify): _________________________

Mitotic Rate
Specify: ___ /5 mm^2
___ Cannot be determined (explain): ____

Note: 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.

Necrosis
___ Not identified
___ Present
    Extent: ___%
___ Cannot be determined

Histologic Grade (Note B)
___ G1: Low grade; mitotic rate ≤5/5 mm^2
___ G2: High grade; mitotic rate >5/5 mm^2
___ GX: Grade cannot be assessed

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

Additional Pathologic Findings
Specify: ____________________________

Ancillary Studies (Note D)
Note: 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)
    ___ Positive
    ___ Negative

___ DOG1 (ANO1)
    ___ Positive
    ___ Negative

___ SDHB
    ___ Intact
    ___ Deficient

___ SDHA
    ___ Intact
    ___ Deficient

___ Pending
___ Other (specify): ____________________________

Molecular Genetic Studies (eg, *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 results: ____________________________
___ Not performed

Prebiopsy Treatment
___ No known prebiopsy therapy
___ Systemic therapy performed (specify type): ____________________________
___ Therapy performed, type not specified
___ Not specified

Treatment Effect (Note E)
___ No known prebiopsy therapy
___ Not identified
___ Present
    Specify percentage of viable tumor: _____%
___ Cannot be determined

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.[1-3] 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.[4]

References

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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 mm^2
G2: High grade; mitotic rate >5/5 mm^2

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 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.

C. 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 assessment.[2-6] 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.[2-6] 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. 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)

Tumor Parameters		Risk of Progressive Disease# (%)			
Mitotic Rate	***Size***	***Gastric***	***Duodenum***	***Jejunum/Ileum***	***Rectum***
≤5 per 5 mm^2	≤2 cm	None (0%)	None (0%)	None (0%)	None (0%)
	>2 - ≤5 cm	Very low (1.9%)	Low (8.3%)	Low (4.3%)	Low (8.5%)
	>5 - ≤10 cm	Low (3.6%)	(Insufficient data)	Moderate (24%)	(Insufficient data)
	>10 cm	Moderate (10%)	High (34%)	High (52%)	High (57%)
>5 per 5 mm^2	≤2 cm	None##	(Insufficient data)	High##	High (54%)
	>2 - ≤5 cm	Moderate (16%)	High (50%)	High (73%)	High (52%)
	>5 - ≤10 cm	High (55%)	(Insufficient data)	High (85%)	(Insufficient data)
	>10 cm	High (86%)	High (86%)	High (90%)	High (71%)

Adapted with permission from Miettinen and Lasota.[5] 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.[2-4,6]

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

References

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D. 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.[1,2] 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%).[3-5] 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 (PDGFRA)* mutation.[6] 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 [4,5]. 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).[7]

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. [8-11] 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.[12,13] Patients with SDH-deficient GIST should be referred to a genetic counselor for appropriate work up.

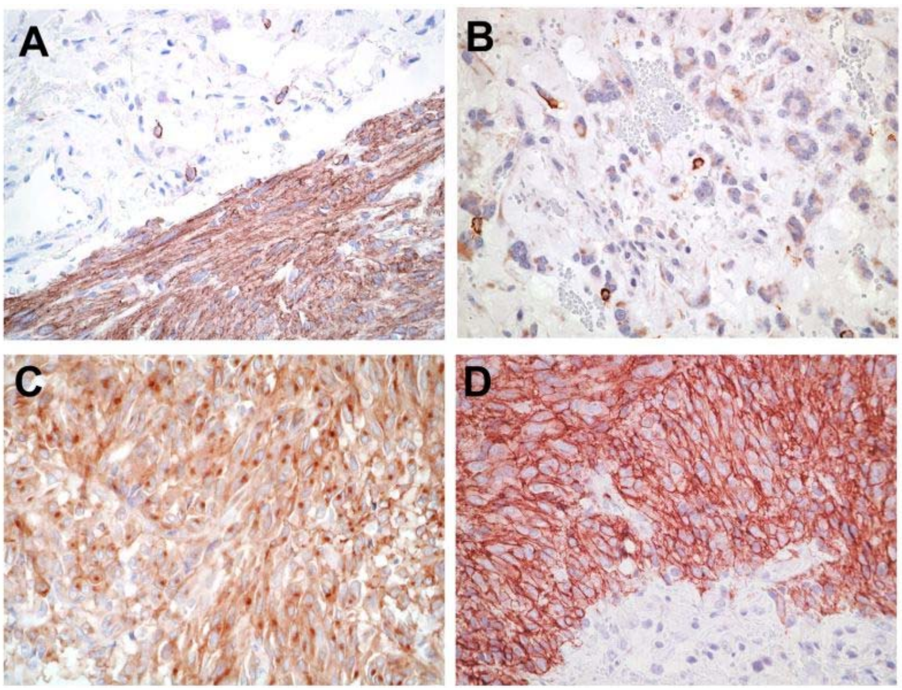

Figure 1. Patterns of KIT staining in gastrointestinal stromal tumor (GIST). A. Diffuse and strong immunoreactivity in a typical GIST. B. Focal and weak pattern in an epithelioid gastric GIST with a *PDGFRA* mutation. C. Dot-like perinuclear staining. D. Membranous pattern. (Original magnification X400.)

Molecular Analysis

Approximately 75% of GISTs possess activating mutations in the *KIT* gene, whereas another 10% have activating mutations in the *PDGFRA* gene.[14-17] 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.[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.[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.

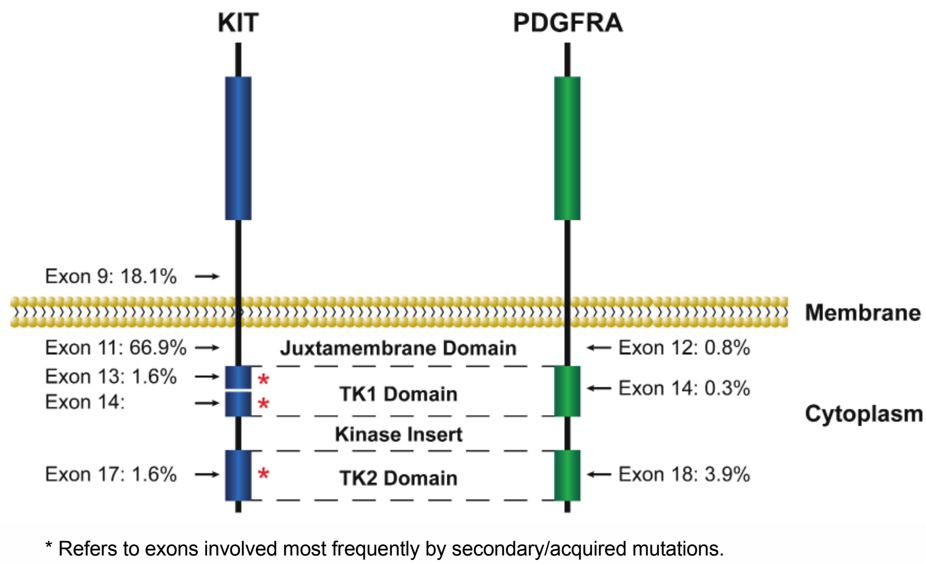

Figure 2. Locations and frequency of activating *KIT* and *PDGFRA* mutations in GIST. Adapted with permission from Heinrich et al.[14] Copyright 2003 by the American Society of Clinical Oncology. All rights reserved.

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.[19-21] 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

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E. 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.