GIST Summit 2010

Diagnosis, Prognosis and *KIT/PDGFRA* Genotyping in Gastrointestinal Stromal Tumors

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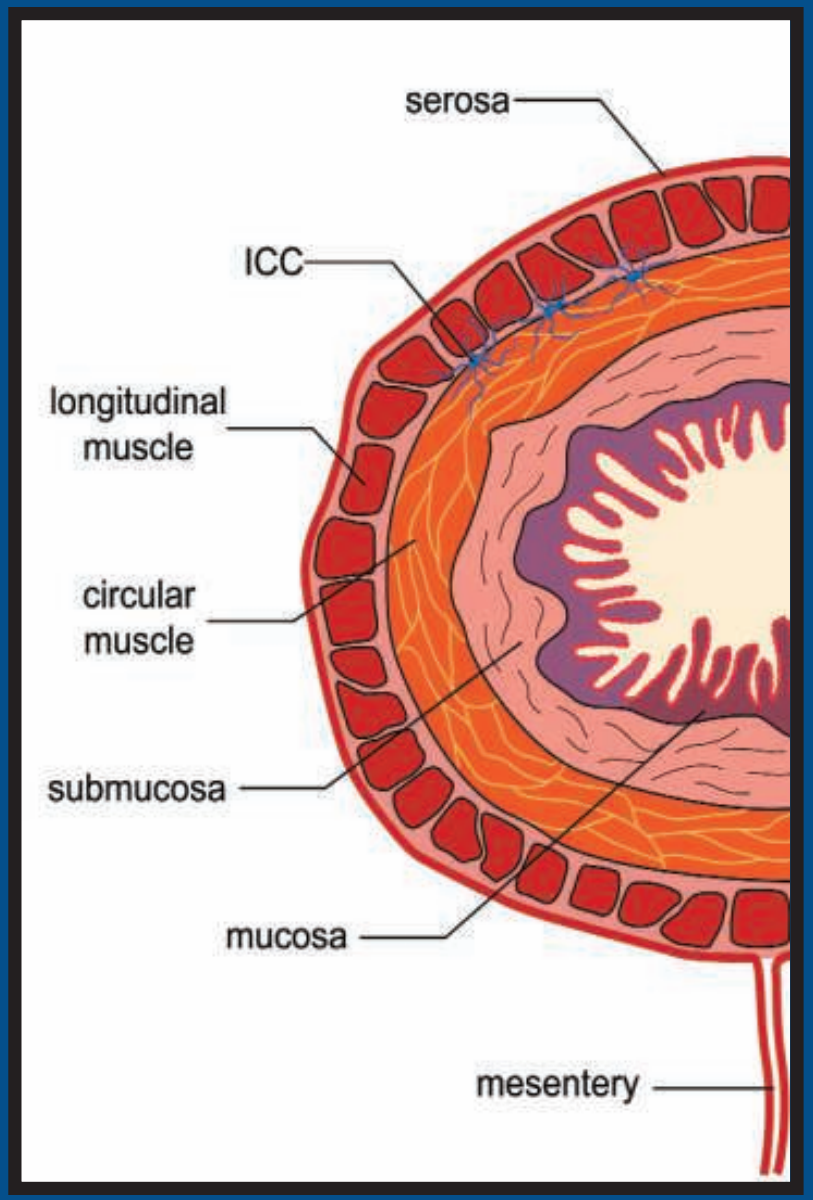
Gastrointestinal Stromal Tumors

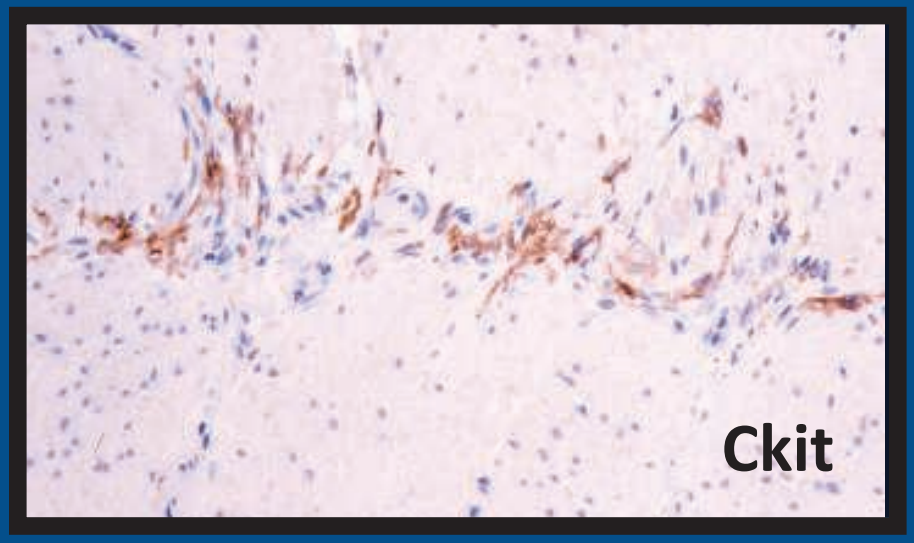
Role of Pathologic Diagnosis and Risk Assessment

Mutation Analysis

Gastrointestinal Stromal Tumor

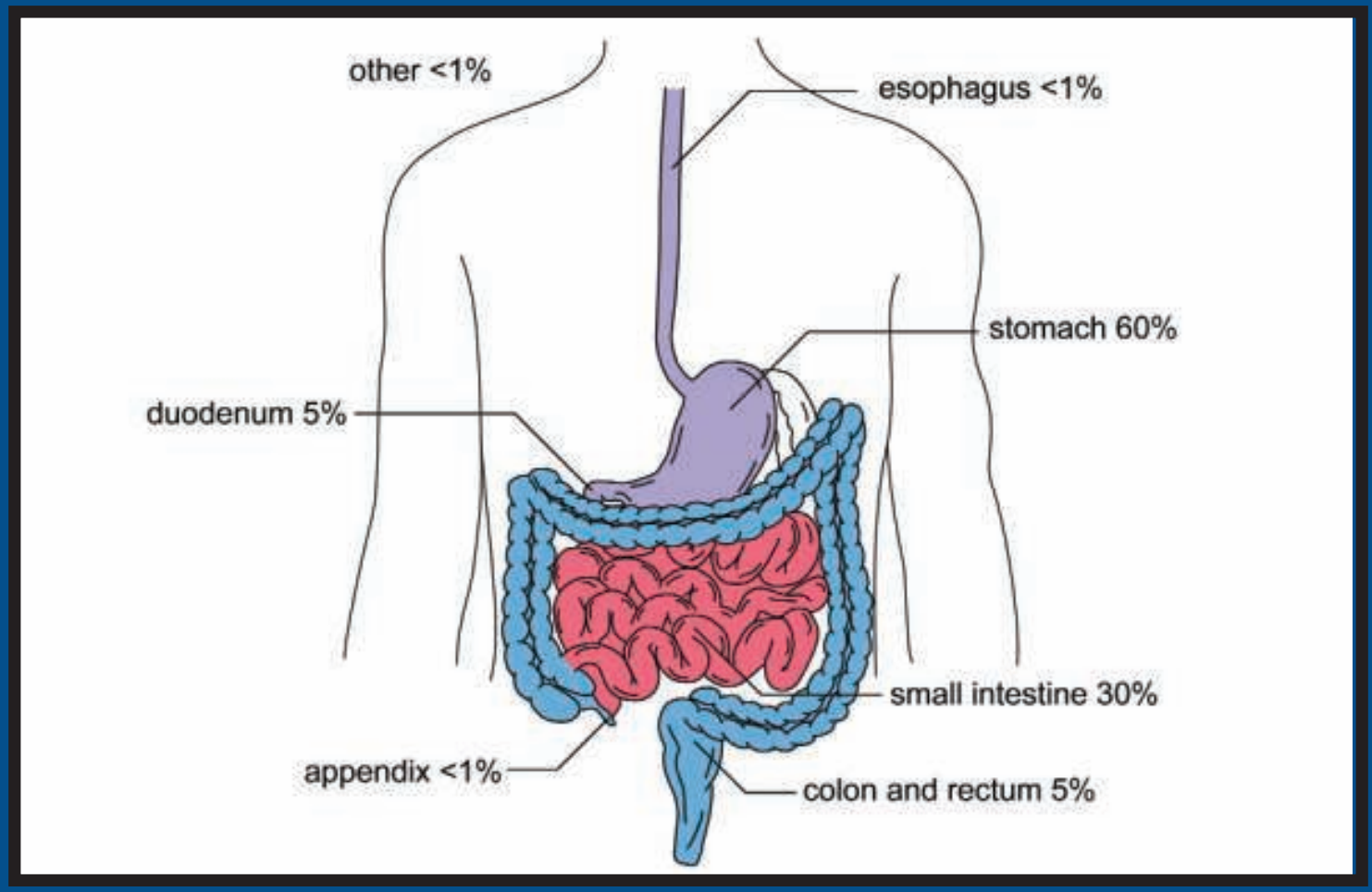


- Arise from the interstitial cells of Cajal (ICC)
- ICC are important in coordinating peristalsis



Courtesy of Brian Rubin

GIST
Sites of Involvement

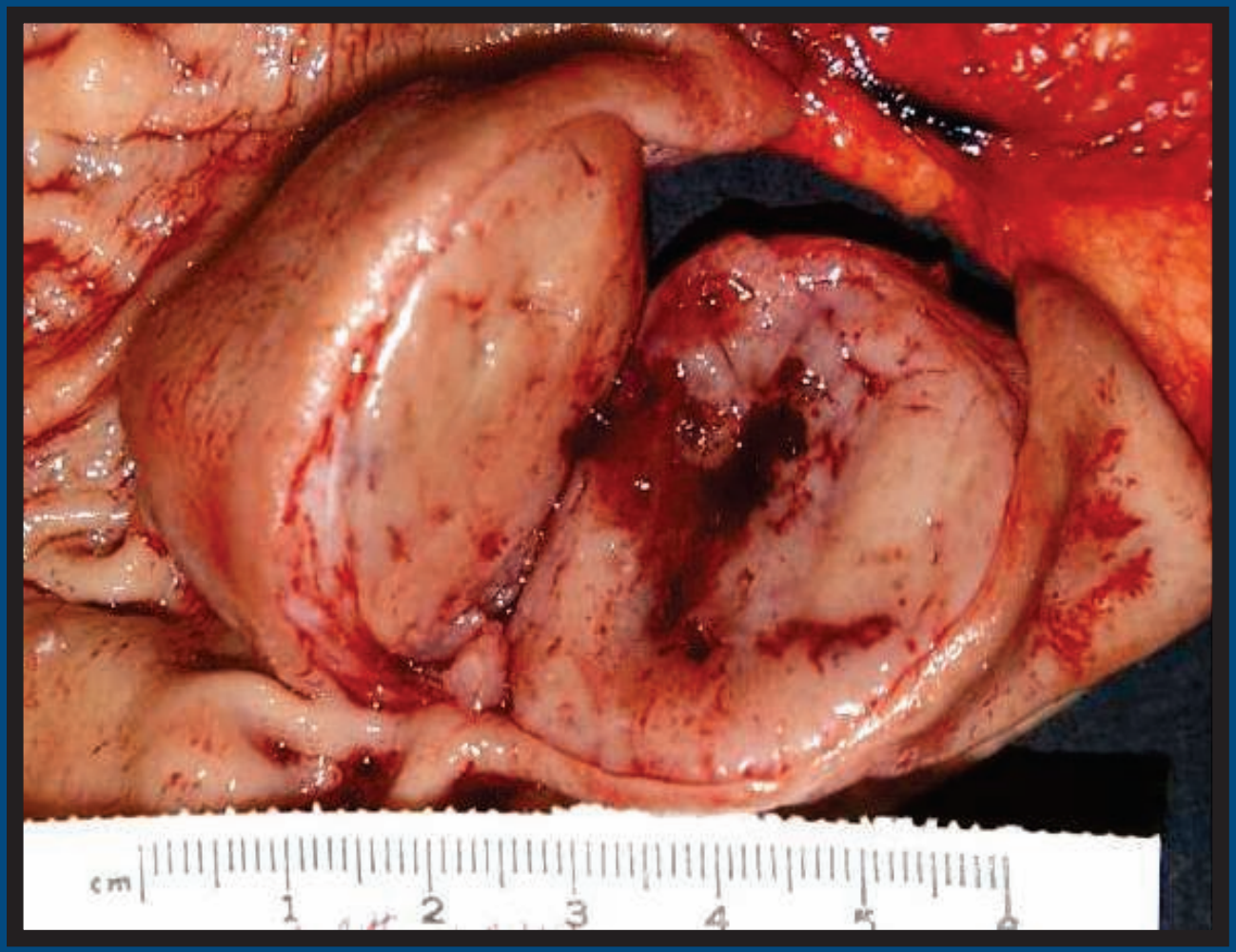


Omentum, mesentery, pelvis and retroperitoneum = EGIST (<1%)

Emory et al. *Am J Surg Pathol*. 1999;23:82.

Gross Appearance

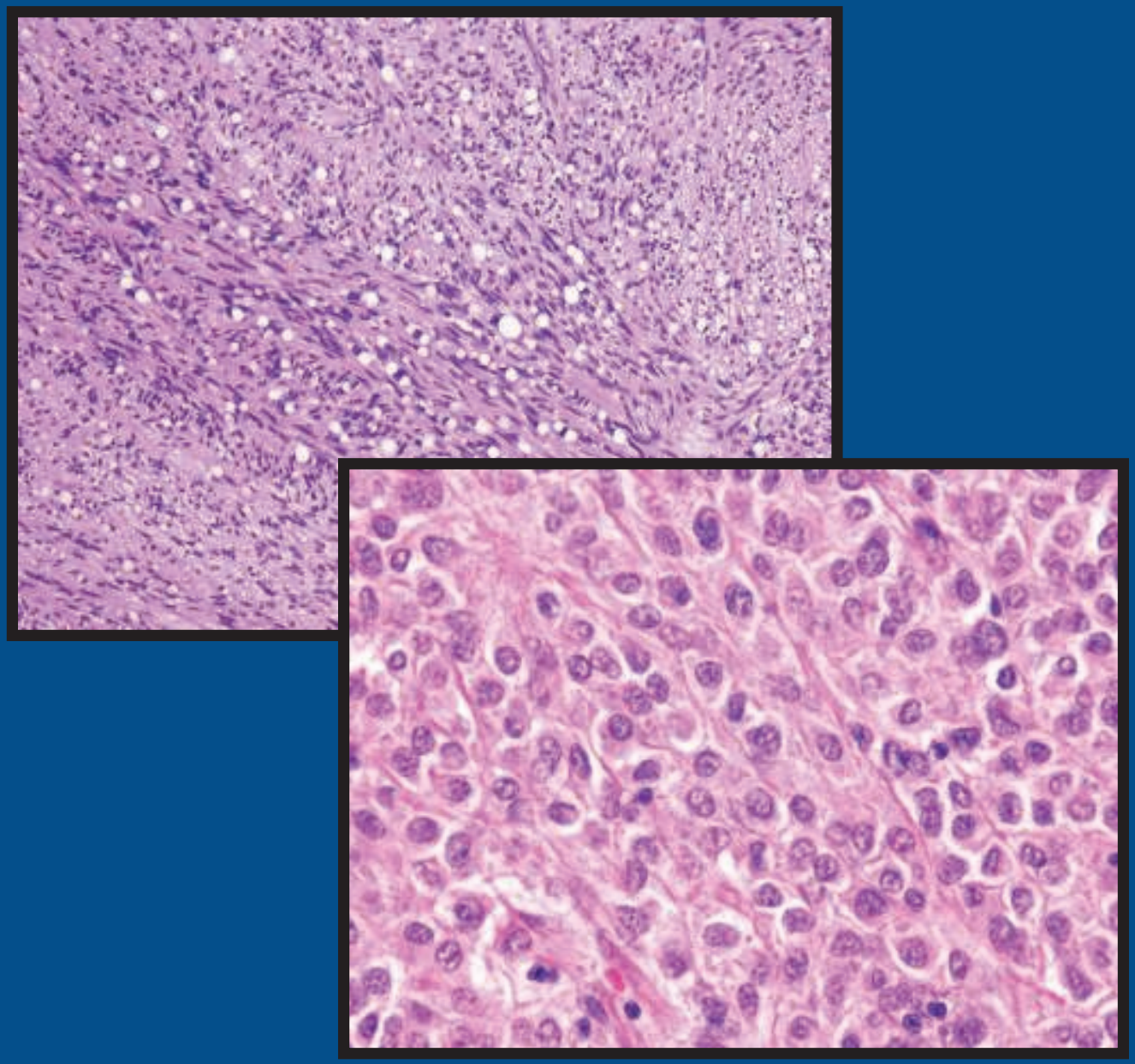
- Most originate from muscularis propria (muscle layers)
- Size varies greatly (median of 10 cm)
- Can grow inwards or out

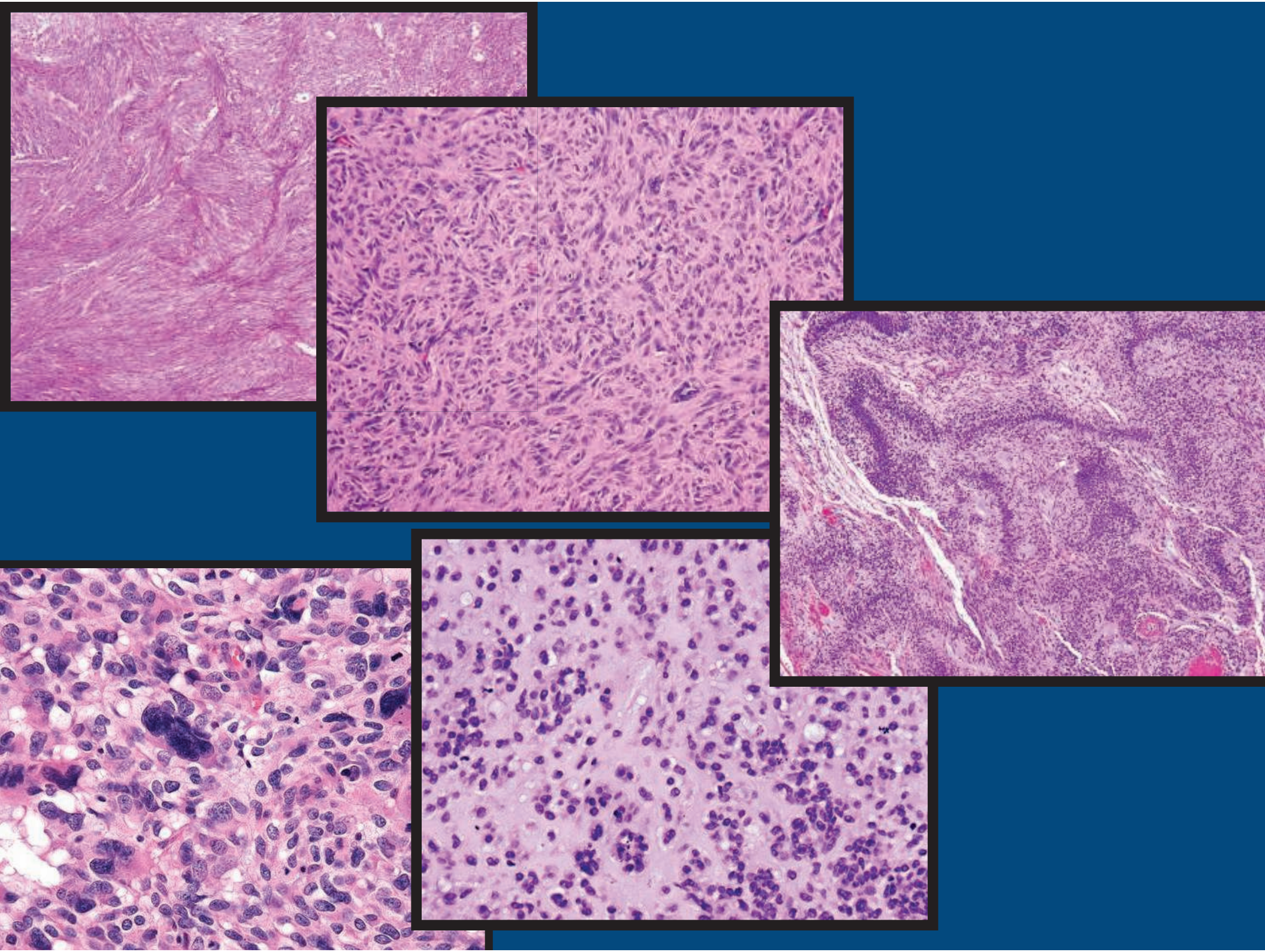


Courtesy of Brian Rubin

GIST
Morphology

- Spindle cell
- Epithelioid
- Mixed





Mimics of GIST

Carcinoma

Melanoma

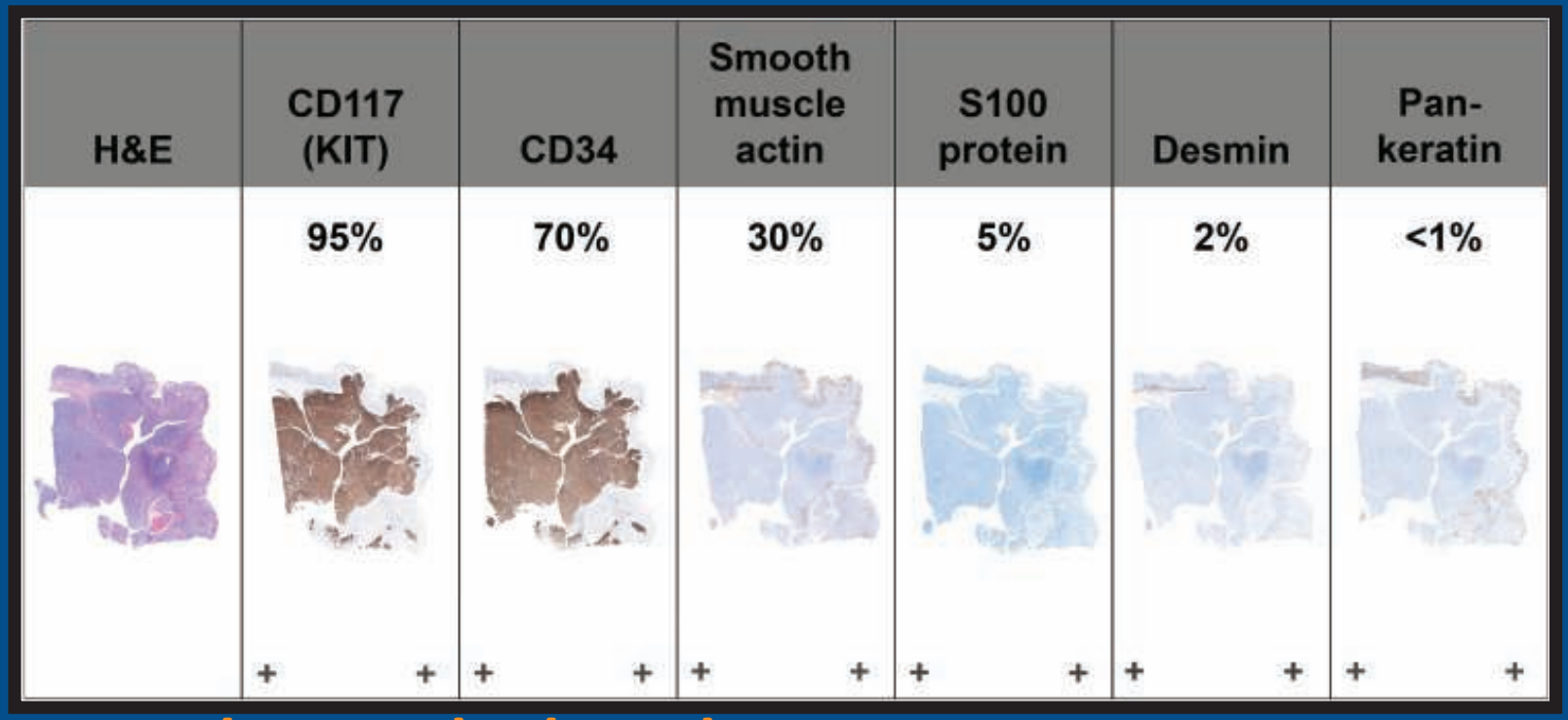
Leiomyoma

Leiomyosarcoma

Schwannoma

Fibromatosis

Immunohistochemical Profile of GISTs



KIT (CD117)+ (95%)

CD34 + (70%)

SMA + (30-40%)

Desmin neg

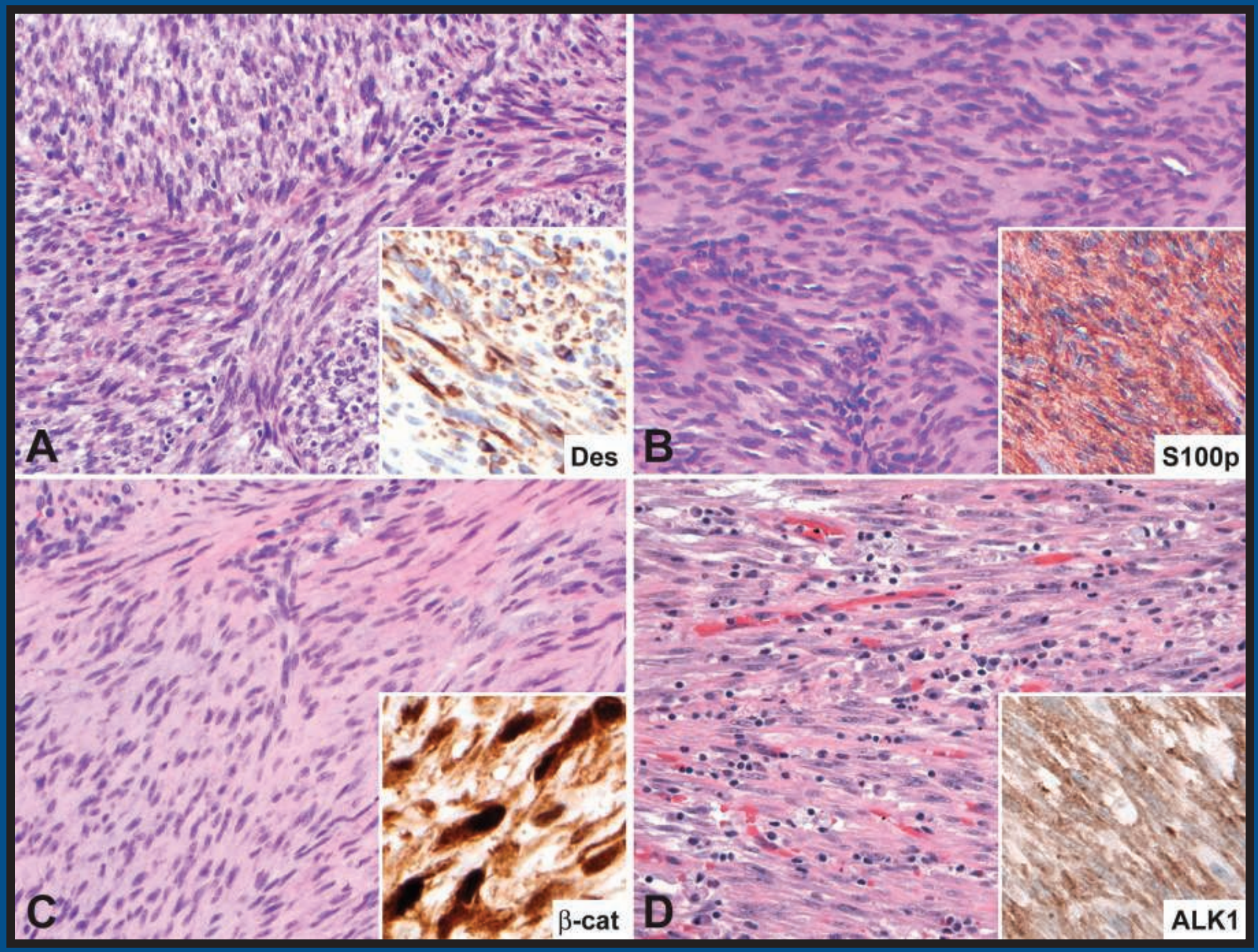
S-100 protein neg

Keratin neg

DOG1 +

(95% / 40% of KIT neg GIST)

Courtesy of Brian Rubin


A
Des
B
S100p
C
β-cat
D
ALK1

GISTs
Clinical Behavior

Behavior is difficult to predict.

Most aggressive GISTs metastasize within 5 yrs.

Small subset may metastasize up to 20 yrs after presentation.

Tendency for intra-abdominal spread and metastasis to liver.

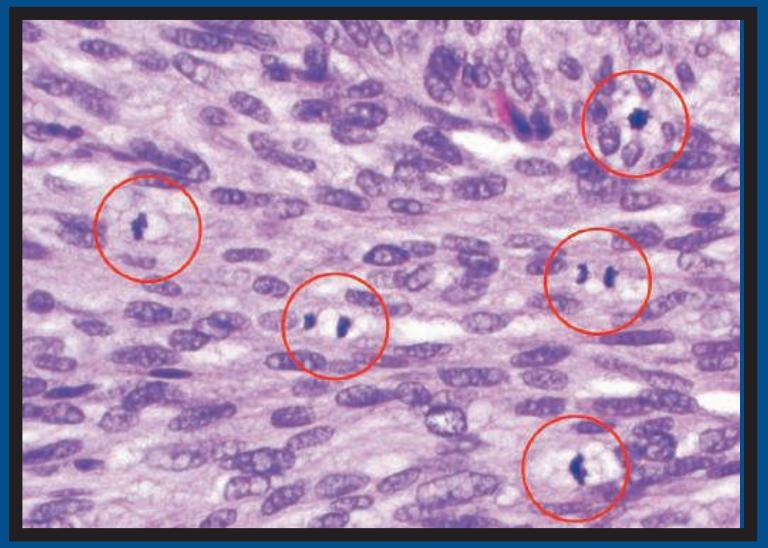
***Never metastasize to lymph nodes.**

Prognostic Factors

Size

Mitotic Rate

Anatomic Location



Pleomorphism

Cellularity

Necrosis

Mucosal Invasion

Proliferation Markers (Ki-67, Mib-1, PCNA, etc)

DNA Flow Cytometry

Image Analysis

Nuclear Organizer Regions

2007 NCCN GIST

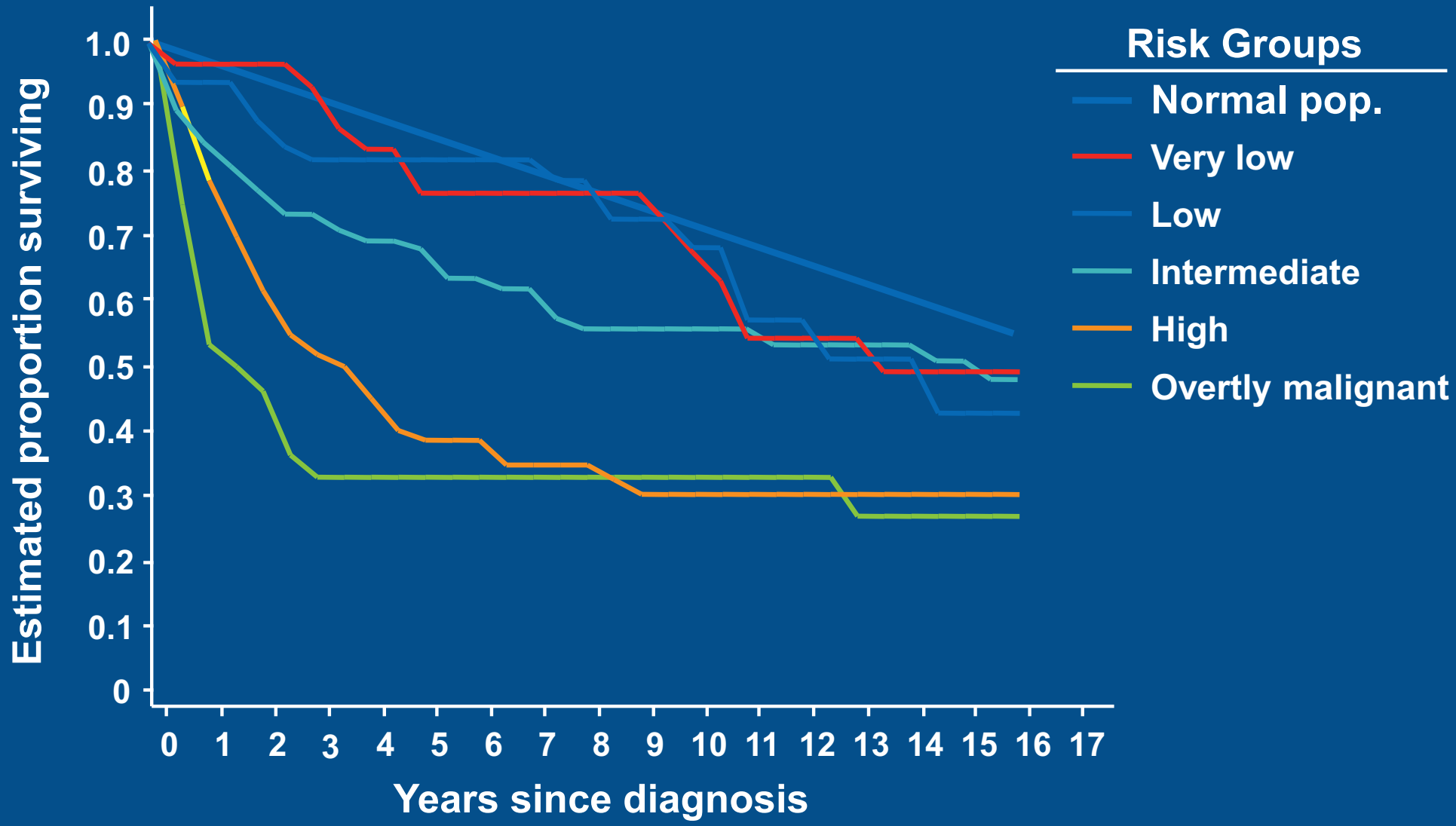
Risk Assessment Guidelines***

Tumor Parameters		Risk of Progressive Disease (%)			
Mitotic Index ≤ 5 per 50 hpf	Size	Gastric	Duodenum	Jejunum / Ileum	Rectum
	≤ 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%)	(Insuff. data)	Moderate (24%)	(Insuff. data)
	> 10 cm	Moderate (10%)	High (34%)	High (52%)	High (57%)
Mitotic Index > 5 per 50 hpf	≤ 2 cm	None*	(Insuff. data)	High*	High (54%)
	> 2 ≤ 5 cm	Moderate (16%)	High (50%)	High (73%)	High (52%)
	> 5 ≤ 10 cm	High (55%)	(Insuff. data)	High (85%)	(Insuff. data)
	> 10 cm	High (86%)	High (86%)	High (90%)	High (71%)

***Modified from Miettinen & Lasota, *Semin Diagn Pathol*, 2006 by Dr. Chris Corless, OHSU.

Data based on long-term follow-up of 1055 gastric, 629 small intestinal, 144 duodenal and 111 rectal GISTs.[Miettinen *et al*. 2005 and 2006]

Overall Survival by Risk Group



Kindblom. At: http://www.asco.org.

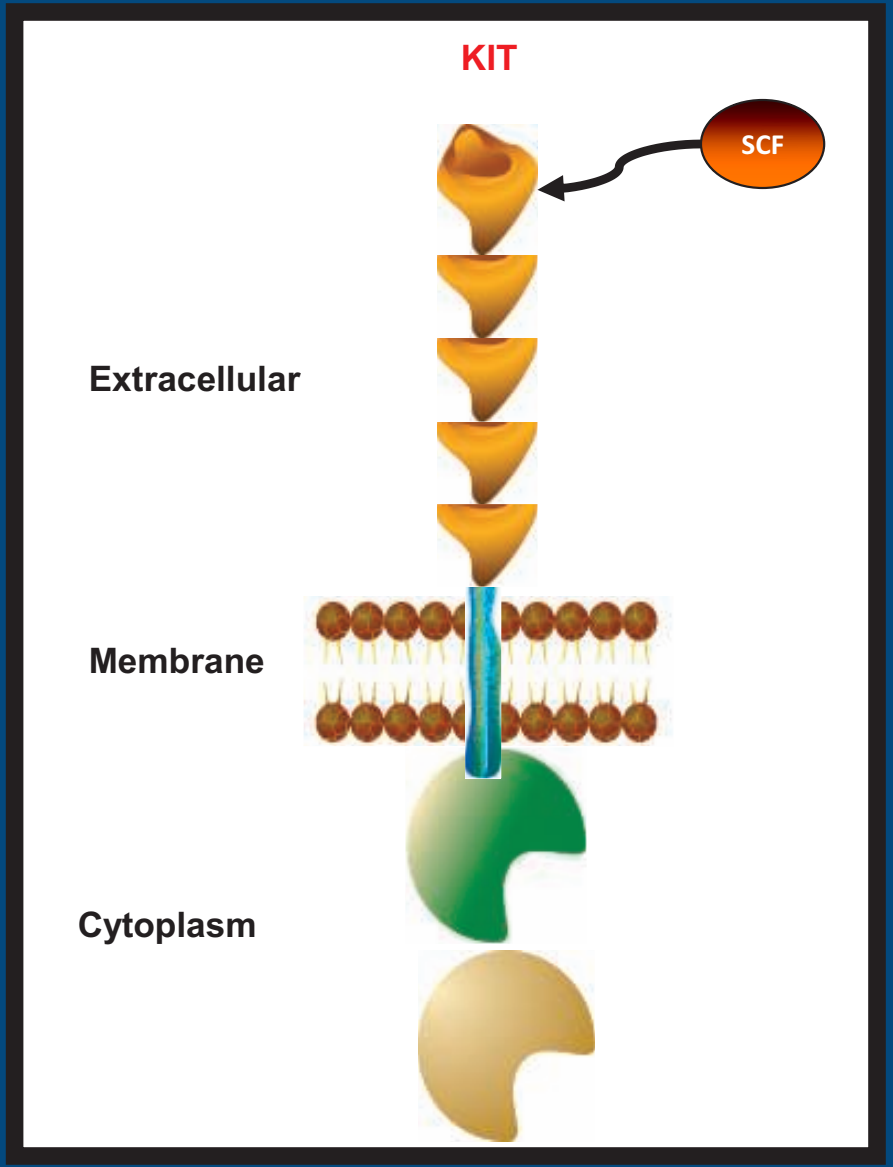
GIST Reporting

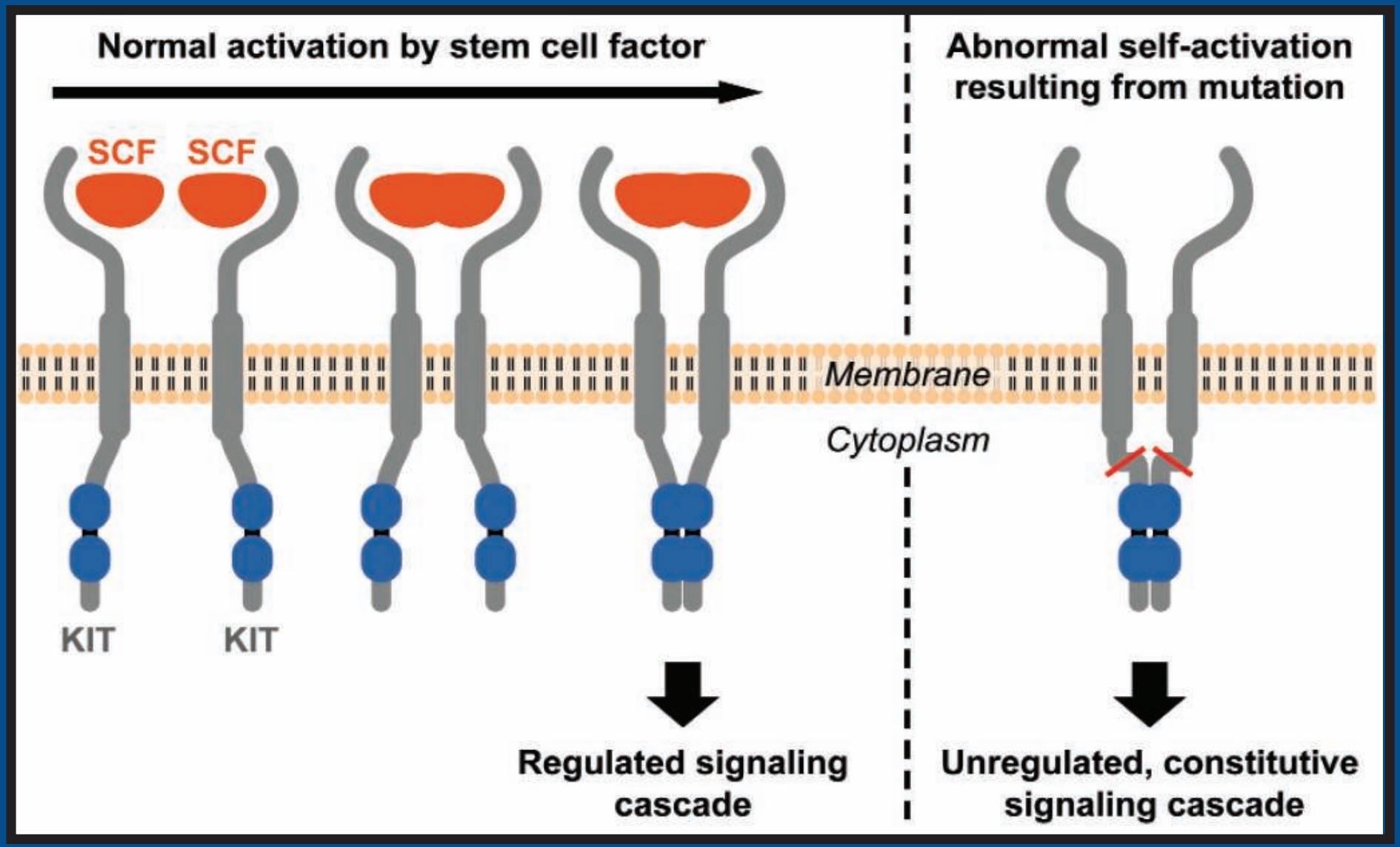
- Size
- Site of Involvement
- Mitotic Count (per 50 hpfs)
- Resection margins
- Document metastases

KIT/PDGFRA Genotyping

GIST

- Majority (86%) of GISTs are characterize with recurrent mutations involving the gene *KIT* or *PDGFRA*
- Both genes encode for proteins which are located on the cell surface
- Plays a role cell growth and survival
- Regulated by a cytokine Stem Cell Factor

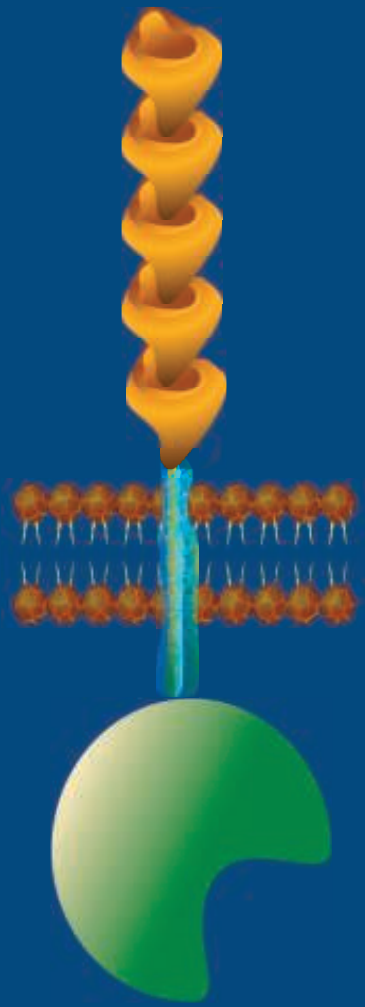



Normal activation by stem cell factor
Abnormal self-activation resulting from mutation
SCF
SCF
Membrane
Cytoplasm
KIT
KIT
Regulated signaling cascade
Unregulated, constitutive signaling cascade

- Most have mutations in KIT
- Certain portion of the genes (i.e. exons) encode for different parts of the protein are characteristically mutated in GISTs
- Beneficial to know which exons are effected

N=950 GISTs Tumors Analyzed In Heinrich & Corless Labs

KIT (78.5%)



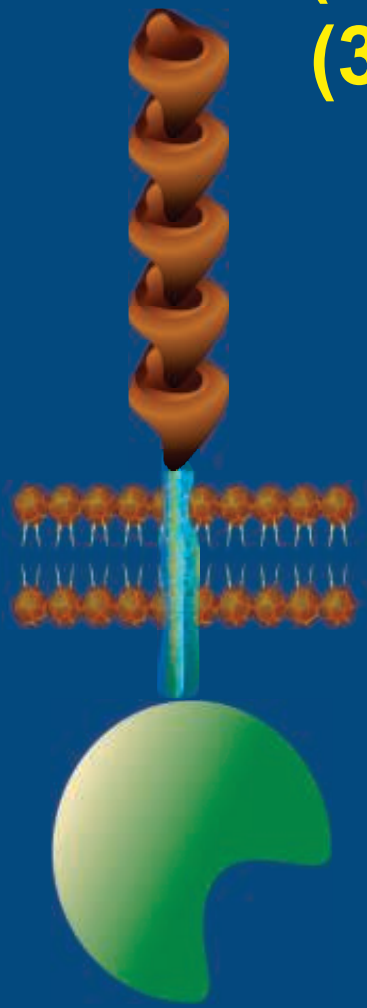
Exon 9

Exon 11

Exon 13

Exon 17

PDGFRA (7.5% total)
(35% of KIT-WT)



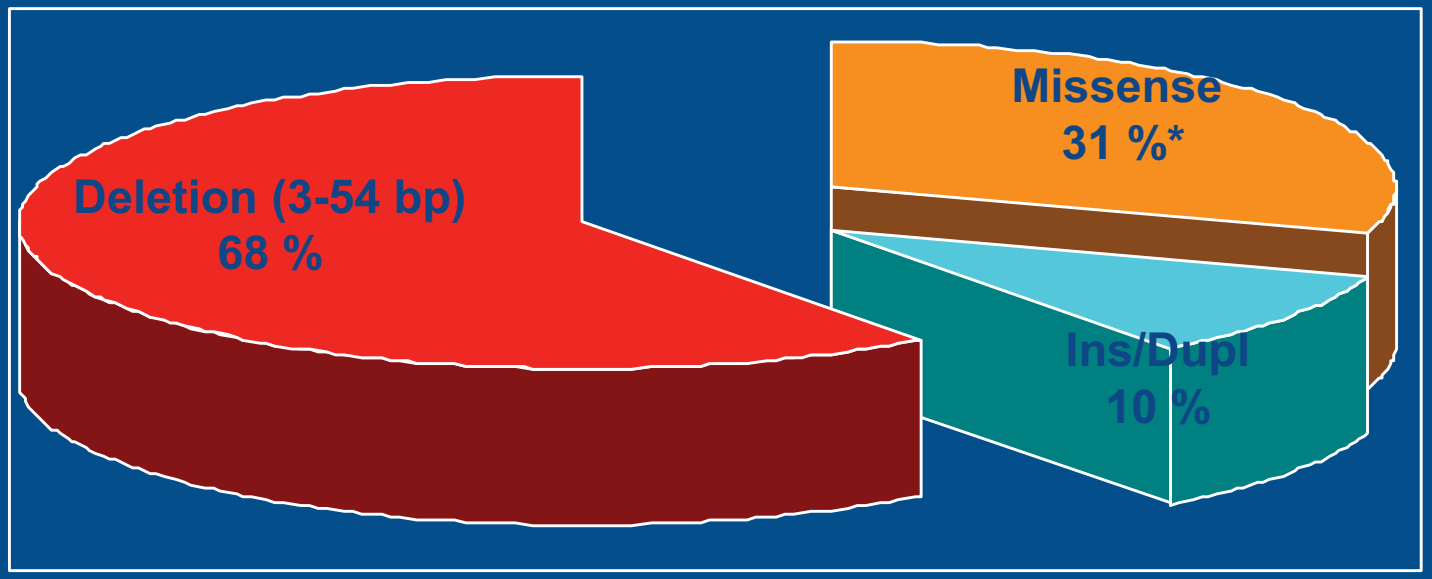
Exon 12

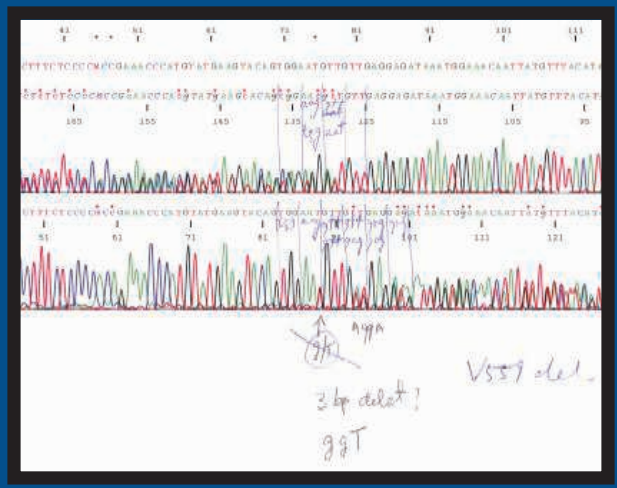
Exon 14

Exon 18

Mutation Types

- Many types of mutations
- Point mutations, deletions, duplications, etc.
- Reported with area of protein effected (ex. V559_V560del, A502_Y503dup, V560D)
- Can also be important to know specific areas of protein involved within an exon
- Area of on-going research

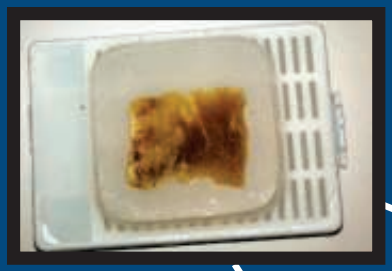




N=58 Exon 11 (UTMDACC)

Analysis of *KIT/PDGFRA* Genotyping

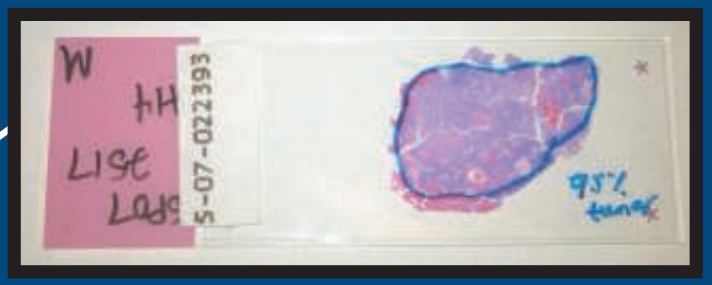
Formalin Fixed Paraffin Embeded (FFPE)



Immunohistochemistry
(CD117, CD34, SMA, Des, S100, Pan-K)

H&E X1 → Review and mark slide for microdissection

Unstained x 10



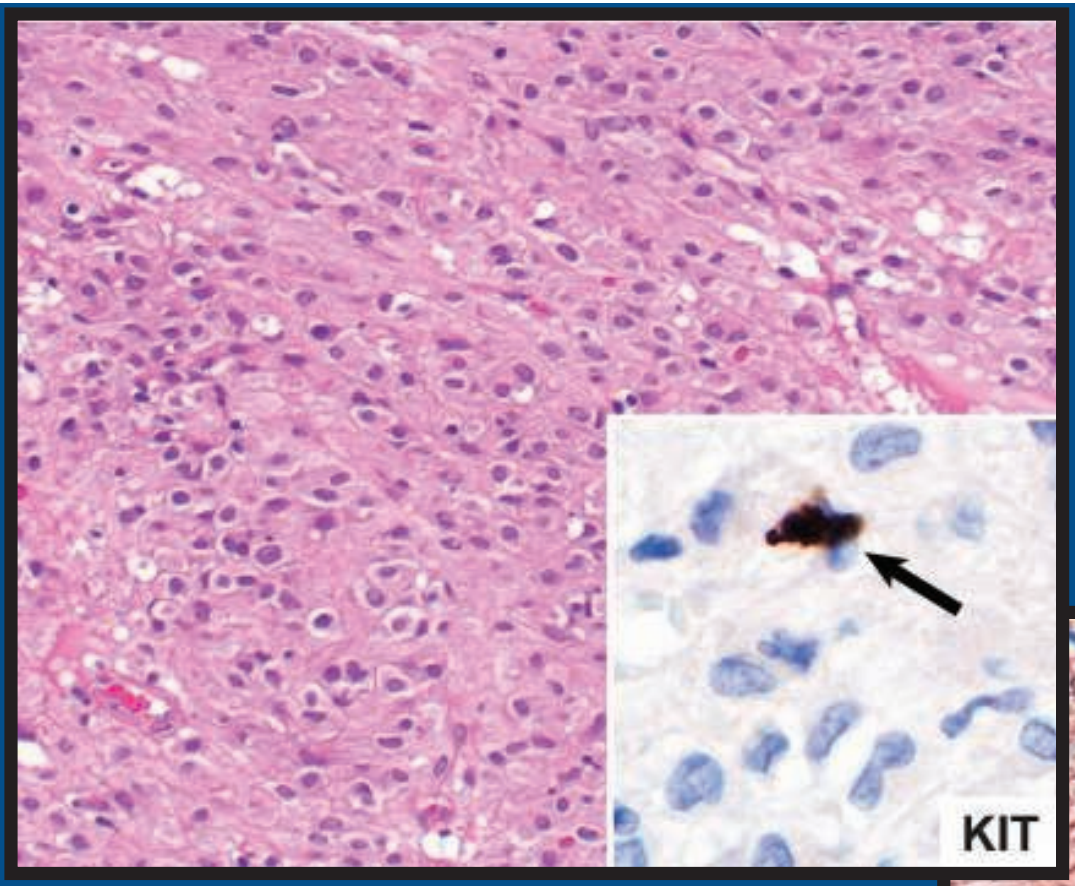
Overlay on H&E and scrape tissue from unstained

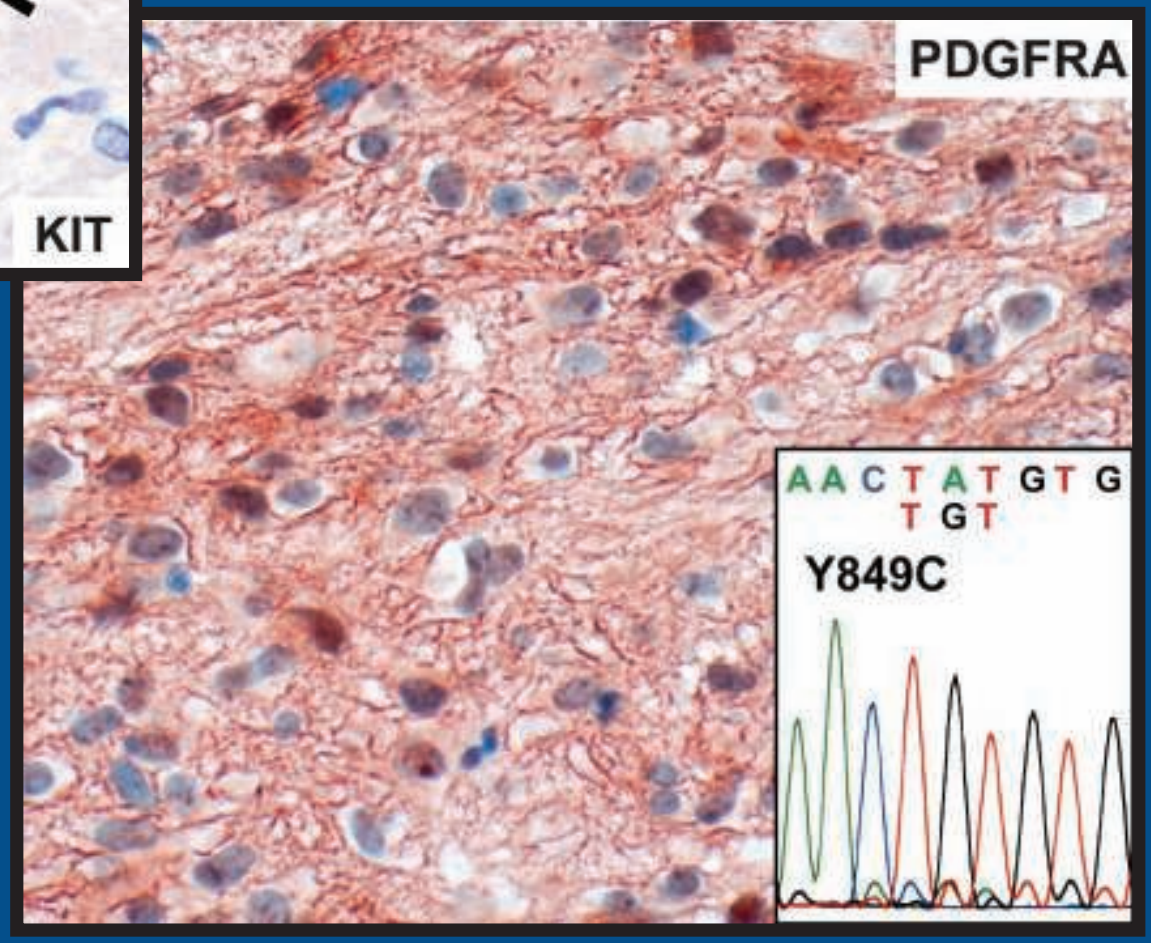
Extract DNA for *KIT* testing (11, 9, 13, 17)
(theoretical 1 in 5 cells – 20 %)

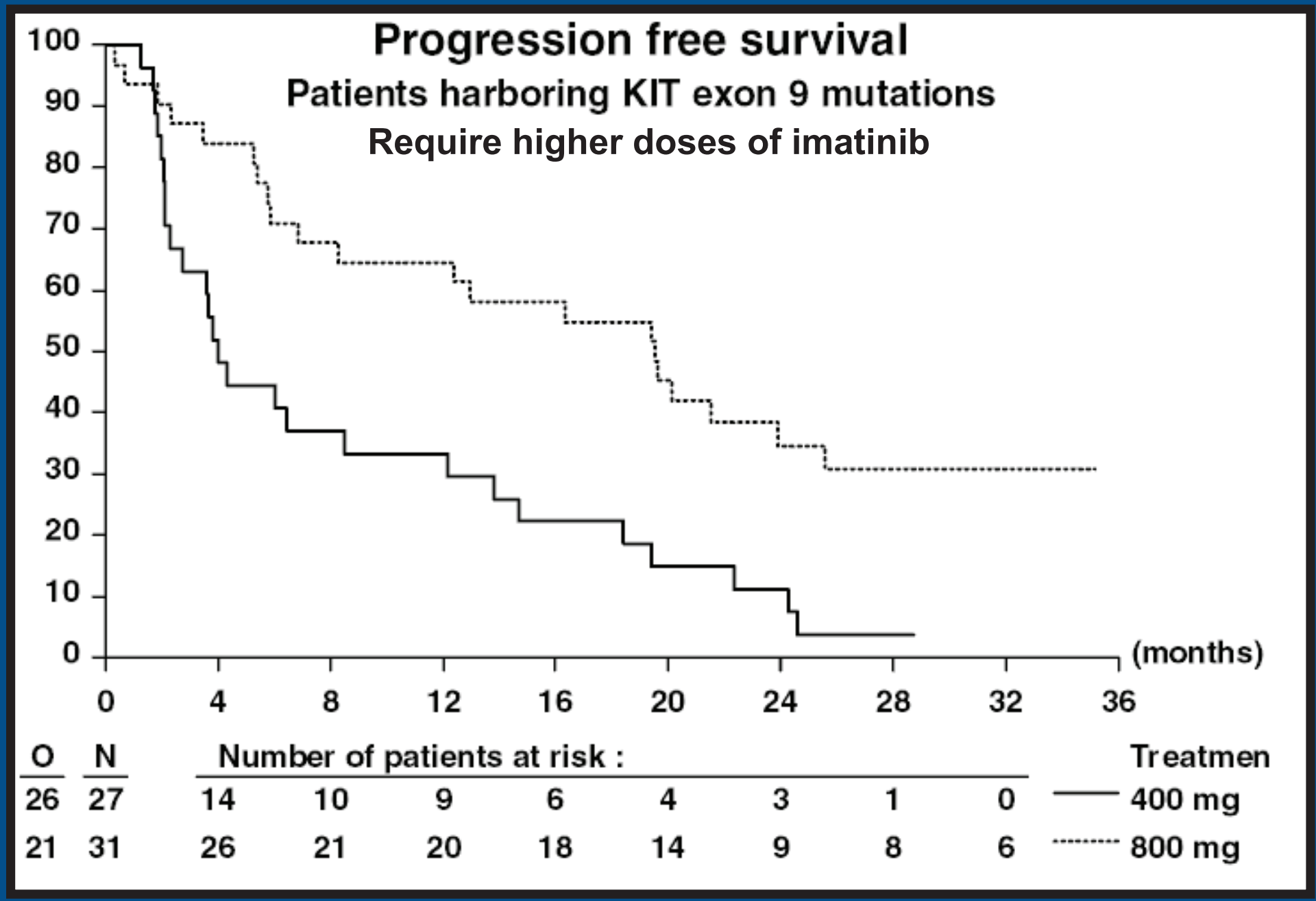
Clinical Use of Kinase Genotyping of GISTs

- **Genotyping of GISTs for *KIT* and *PDGFRA* mutations may be useful for:**
  1. **Confirmation of diagnosis KIT IHC (-) GISTs**
  2. **Prediction of clinical response to imatinib and those that may require different dosage of imatinib**
     - **ex. exon 9**
  3. **Triage patients who are at high risk of failing imatinib therapy to enroll in other clinical trials**
     - **ex. PDGFRA D842V mutations and Wildtype**

Confirmation of Diagnosis in KIT IHC (-) Tumors







Debiec-Rychter, *et al*. Eur J Cancer, 2006.

Frequency and Clinical Significance of *KIT*

KIT exon 11: (60–70%) Arise anywhere in GI tract. Most responsive to Imatinib.

KIT exon 9: (5–15%) Small intestine. Respond to Imantinib at higher doses.

KIT exon 13: (1%) Clinical responses to IM observed but uncharacterized. (***IM resistance point mutations)

KIT exon 17: (1%) Clinical responses to IM observed but uncharacterized. (***IM resistance point mutations)

Frequency and Clinical Significance of PDGFRA and being WT

PDGFRA exon 12: (1%) Rarely originate from the intestine. Clinical responses to IM observed.

PDGFRA exon 14: (<1%) Unknown, only few tumors described in the literature.

PDGFRA exon 18: (5%) Most originate from the stomach. D842V Resistant to IM.

Wild type: (10–15%) Primary resistance to imatinib more common; 40% respond to IM.

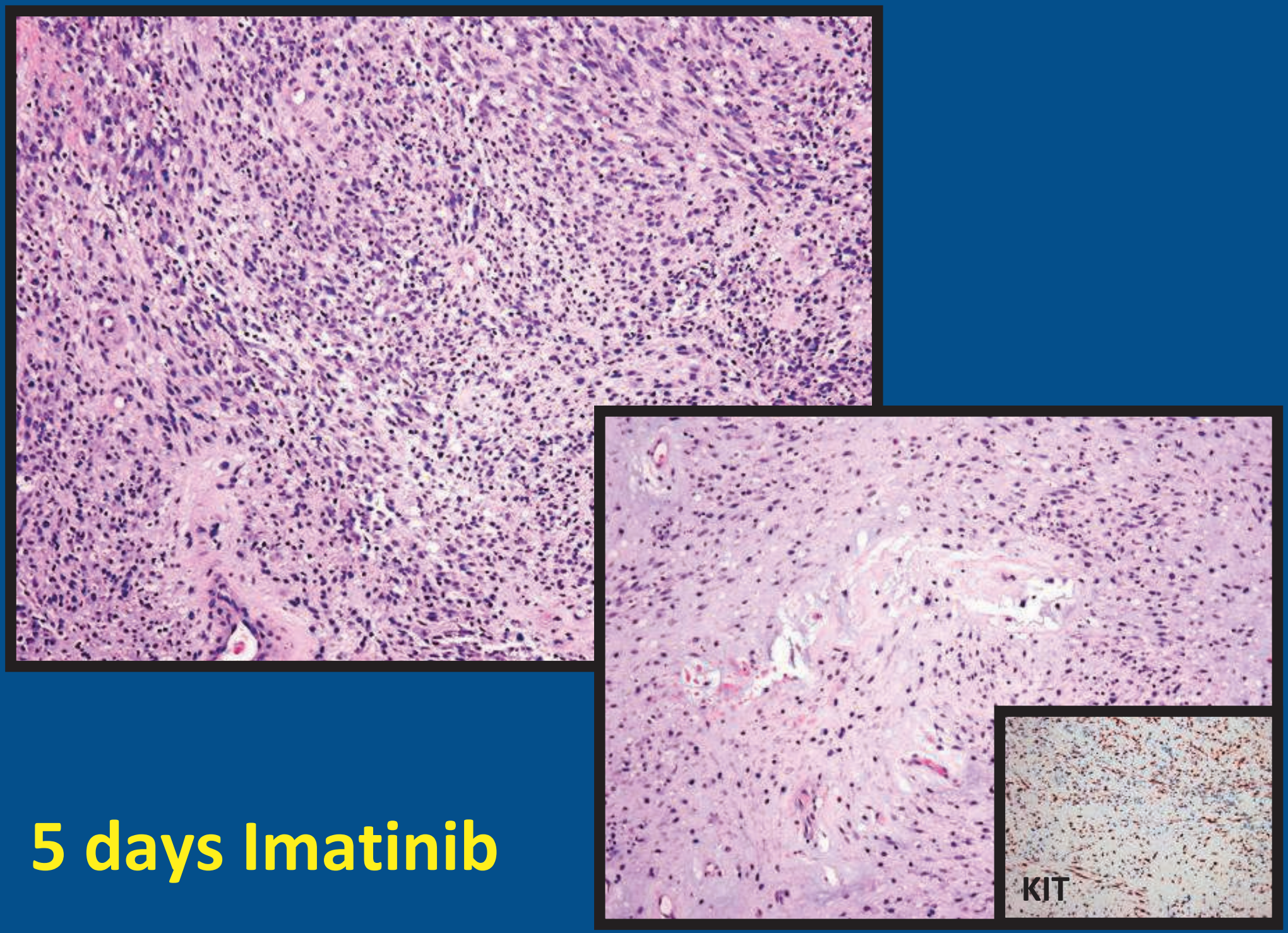
Role of KIT genotyping and Resistance

- **Primary Resistance :** 10-15% will not response as well to imatinib – intrinsically resistant

  - ex. Wildtype (indicate other mechanisms involved), PDGFRA D842V

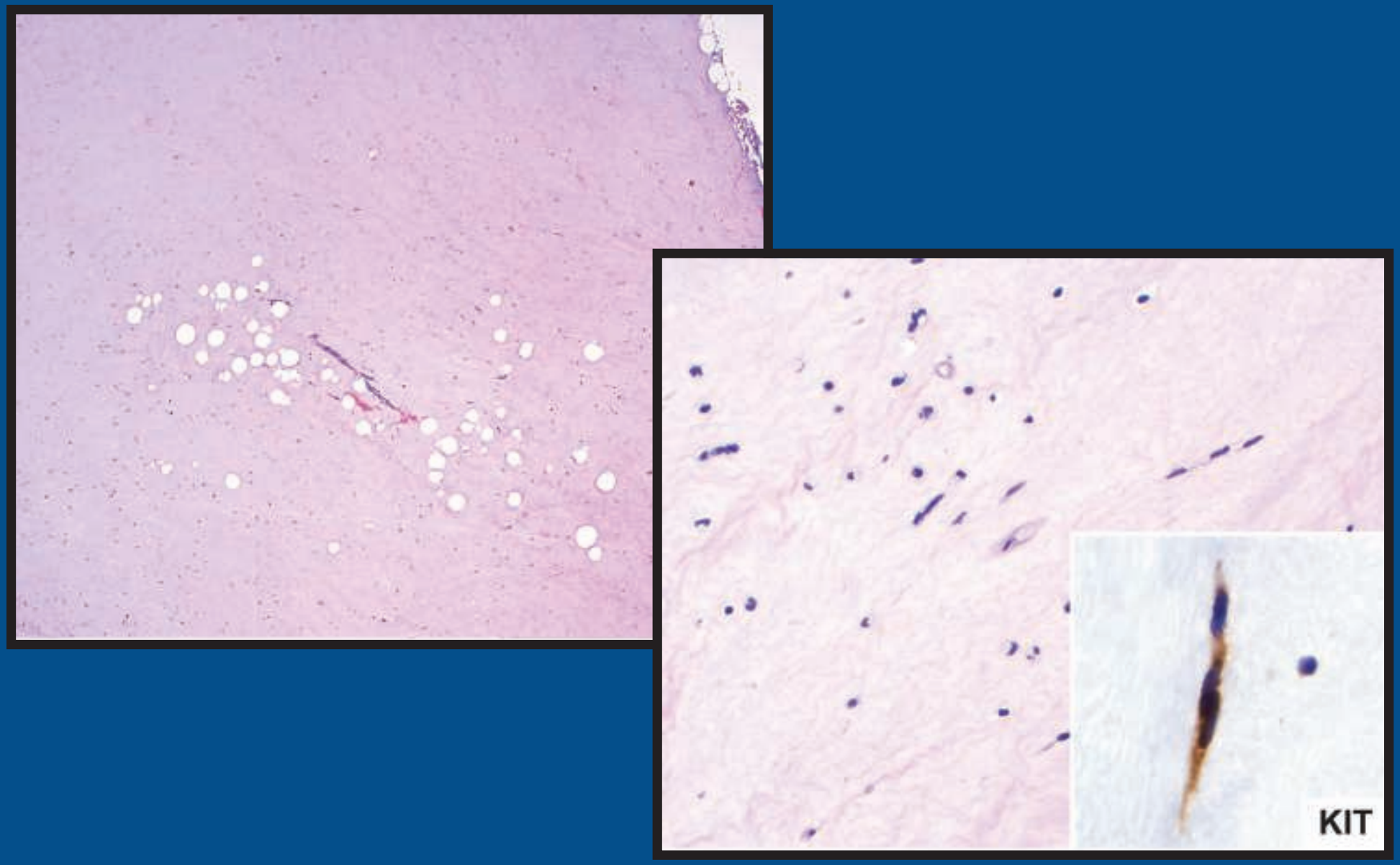
- **Secondary Resistance :** 50-70% of patients on imatinib will progress and develop resistance

- Most common cause is the development of a second mutation
- Most often involve exons 13 and 17 effecting the activation A loop or ATP binding domain resulting in shift to active confirmation or blocking imatinib binding



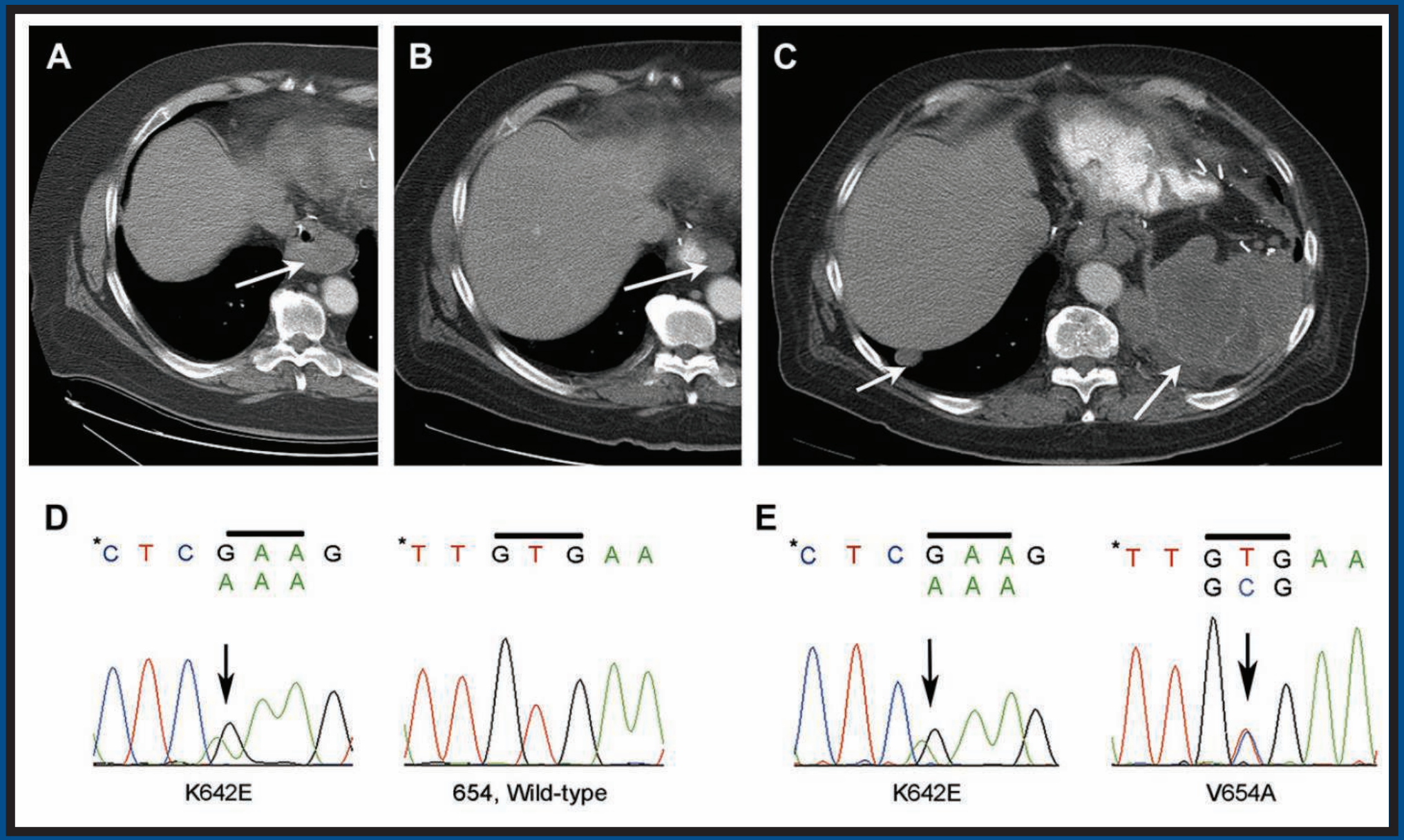
5 days Imatinib

Long term Imatinib Treatment



Secondary Resistance

- In advancing disease, multiple clones can exist within the same tumor nodule and within separate tumor nodules; each with different mutations
- Very heterogeneous
- Mutation testing does not typically need to be done for secondary resistance



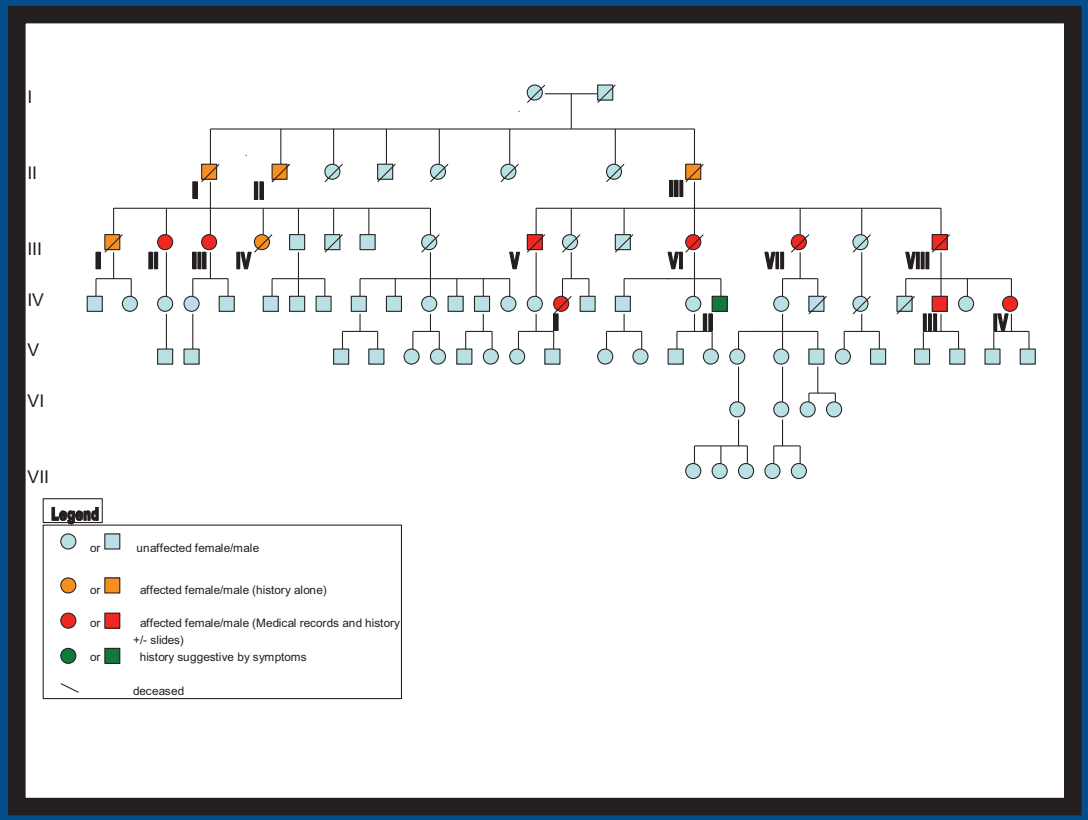
McAuliffe JC et al. Mol Oncol. 2008 2(2):161-3.

Other Mechanisms of Resistance

- KIT Amplification
- BRAF mutations (KIT Wildtype)
- Insulin Growth Factor
- Loss of Heterozygosity
- AXL overexpression
- Heat Shock Protein 90
- Decreased absorption of imatinib

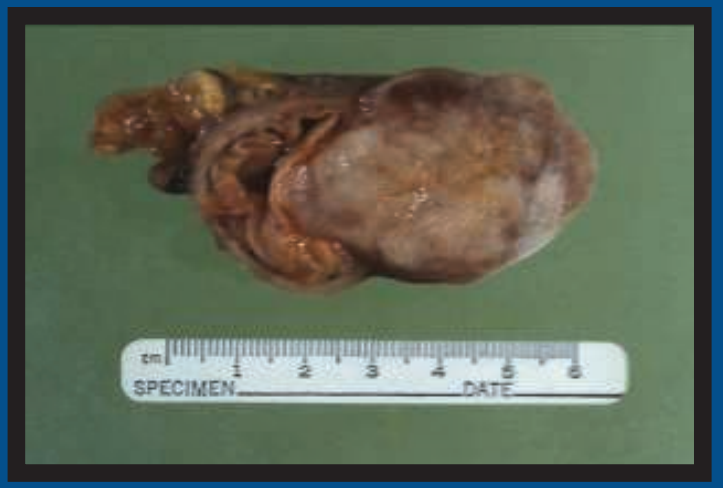
Familial GIST

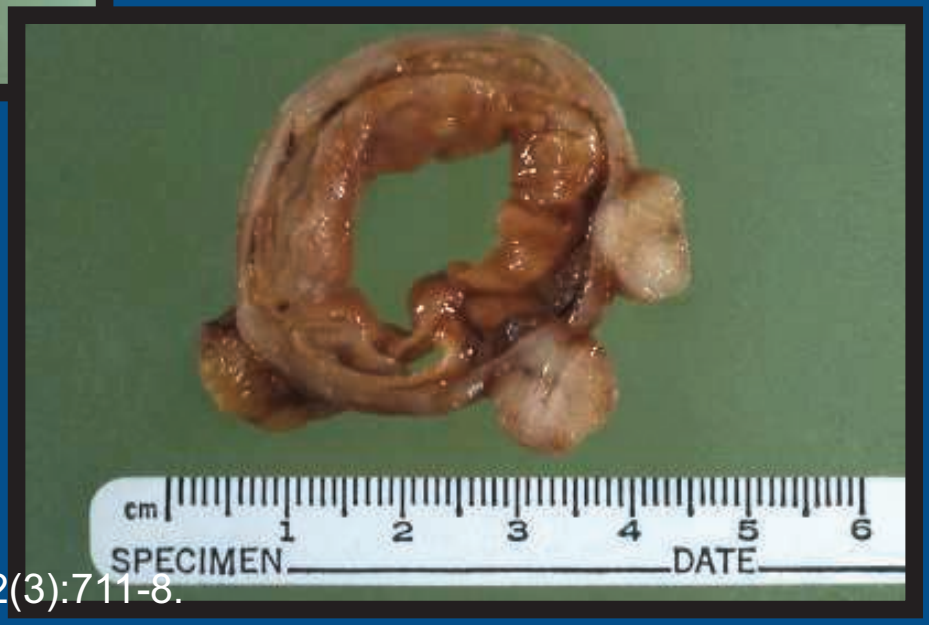
- **Germline mutation in exon 11.**



Kleinbaum EP et al. Int J Cancer. 2008 Feb 1;122(3):711-8.

Familial GIST
Gross Pathology





Kleinbaum EP et al. Int J Cancer. 2008 Feb 1;122(3):711-8.

GISTs associated with Neurofibromatosis

- Similar morphology.
- Mutations are different.
- Wild type for *KIT*
- Different mechanism for these tumors.
- IGFR inhibitors?

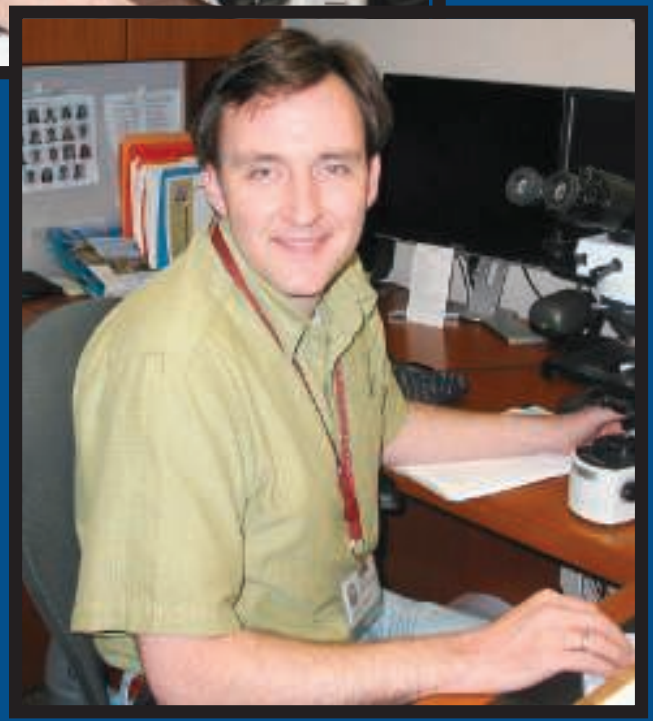
Thank You.

- Dr. Alexander Lazar MD/PhD
- Dr. Jonathan Trent MD/PhD

http://www.gistsupport.org/for-new-gist-patients/understanding-your-pathology-report-for-gist.php

Gastrointestinal Stromal Tumor

Understanding Your GIST Pathology Report



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