Supplemental Data

Imaging, image analyses

Imaging

FDG-PET imaging was performed after fasting for at least 4 hr. Patients had blood glucose measurement and were then injected with 10-20 mCi FDG intravenously. Scanning from the base of the skull to the proximal thighs was performed beginning approximately one hour later, following emptying of the bladder. Approximately 49% of the studies were obtained with dedicated PET scanners, and the remainders were performed with integrated PET/CT scanners. For those patients imaged on dedicated PET scanner, Ge-68 PET transmission scanning matching the areas covered by the emission scan was performed for attenuation correction of the emission scan. Helical CT imaging was performed over the same anatomic range of the PET emission scanning for patients imaged on PET/CT scanners. CT-based attenuation correction was performed on patients imaged with PET/CT scanners.

Image analyses

The FDG-PET scans were interpreted by each of two independent readers blinded to other clinical information. A 5-point ordinal scale was used for visual comparison of the pre- and post-treatment FDG uptake in tumor foci as follows: marked decrease, slight decrease, no change, slight increase, marked increase. Once this reading was completed, the scans were then re-read by each reader with knowledge of the conventional cross-sectional imaging findings and clinical

information followed by a consensus reading at which time target lesions were defined.

Masking regions of interest (ROIs) were placed around up to three target lesions as determined by visual inspection, and placed in the axial plane containing the SUV_{max} for that tumor site. A computed-defined ROI was then determined based on a 70% maximum uptake isocontour. Four background ROIs were also drawn for each tumor by placing copies of the ROI used to measure the tumor in normal-appearing regions of the tissue/organ of origin on the same slice used for tumor analysis. In the absence of comparable normal tissue in the proximity of the tumor, background ROIs were placed on the contralateral side of the body. Background subtraction was used at the time of the design of this National Cancer Institute-approved protocol to correct for the contribution of physiologic uptake in normal parenchyma, to derive target-to-background (TBR) ratios, and to compare these to SUV_{max} and SUV_{mean} in each ROI. Of note, most current quantitative data analysis approaches no longer include background subtraction techniques. These data were originally collected prior to the standardization of semi-quantitative techniques in PET analysis that are available today. The TBR was computed for each lesion by calculating the ratios of the mean SUVs for the tumor and background regions. For each tumor, the SUV_{max} and mean SUV within the 70% isocontour (SUV_{mean}) were recorded, along with the number of pixels. If there was more than one tumor, the SUV_{max} for all lesions was summed and background-subtracted SUVs were calculated by subtracting the sum of

SUV_{max} for the background ROIs from the summed SUV_{max} for all lesions. After correction for radioactive decay, SUVs were computed according to the following:

$$SUV = \frac{\text{ROI activity } (mCi/mL) \times \text{body weight } (g)}{\text{Injected dose } (mCi)}$$

This ROI analysis was performed by another physician (YM or ASB) who could use conventional cross-sectional imaging information to help draw the regions of interest. For the post-treatment measurements, if the tumor had elevated FDG uptake compared to background, ROIs were drawn in the axial plane containing the SUV_{max} for that tumor site using the 70% maximum uptake isocontour as was done at baseline. If the tumor did not have elevated FDG uptake compared to background, the ROI was drawn around the observer's best estimate of the tumor margin. In this case, information from cross-sectional anatomical images and from the previous FDG image could be used to guide the ROI placement.

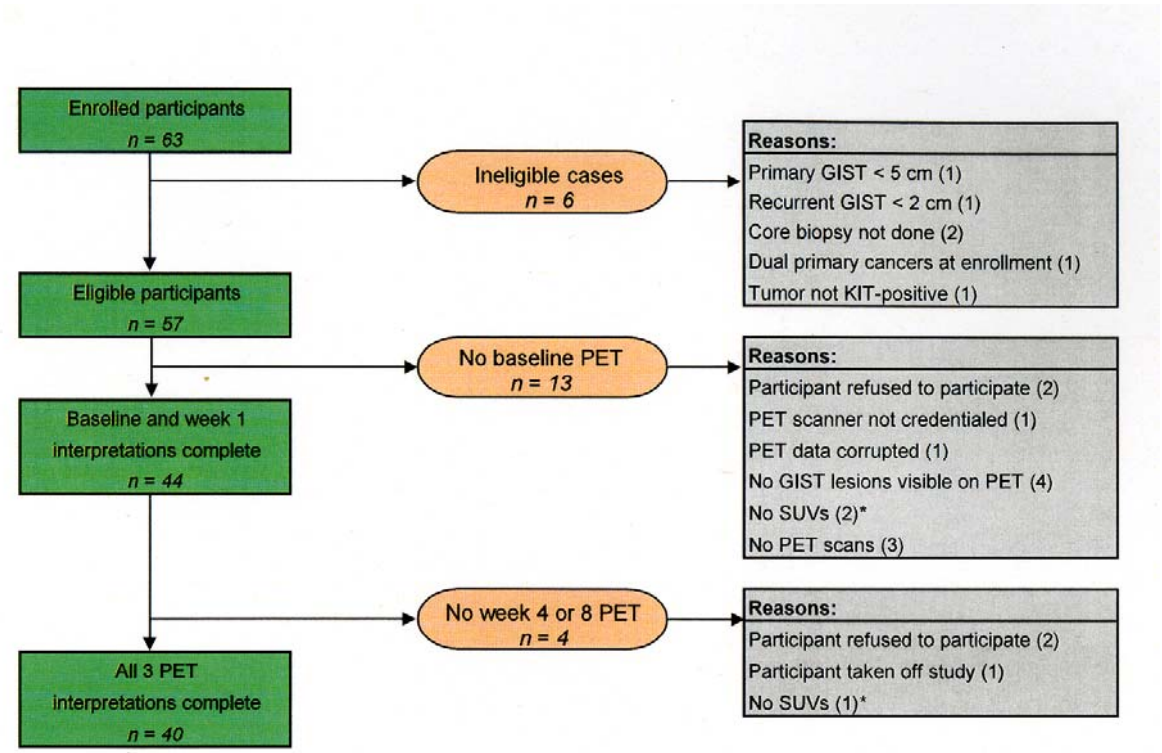
Tumor tissue analyses

Clinical specimens

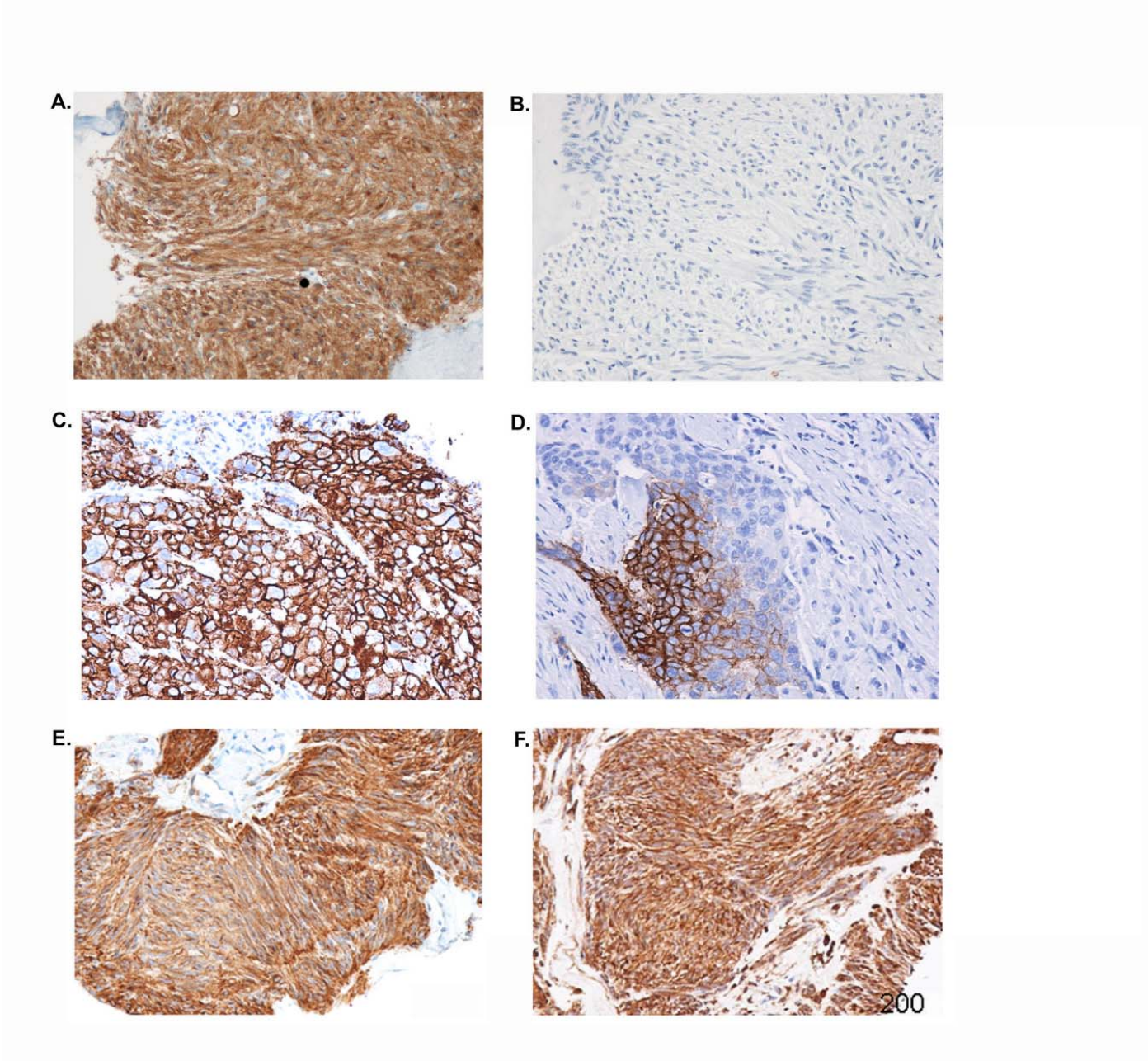
IHC staining was performed on 5-mm sections of paraffin-embedded pre-treatment (n=22) and post-treatment tumor samples (n=22). After deparaffinization and rehydration, sections were subjected to heat-induced epitope retrieval by immersion in a 0.01 M citrate buffer (pH 6.0). Endogenous peroxidase activity was blocked for 15 min in 3% hydrogen peroxidase in methanol. Nonspecific binding was blocked by treatment with a blocking reagent (protein block serum-free; Dako) for 30 min at room temperature. Slides were

then incubated overnight with GLUT1 (Thermo Scientific) or GLUT4 primary antibody (Millipore), diluted to a concentration of1:1000 and 1:500 respectively, at 4^{o} in a humidified chamber. Immunodetection was performed with the DakoEnVision^{TM+} system. The antigen-antibody immunoreaction was visualized using 3–3'-diaminobenzidine as the chromogen. The negative control was performed by replacing the primary antibody with normal horse serum. Sections were examined by an observer blinded to the tumor's response to IM. Images were recorded with a Nikon Eclipse E600 DS-Fi1 digital microscope camera. IHC results were assessed without prior knowledge of clinical findings or treatment. The immunoreactivity was semiquantitatively scored using a well-established immunoreactivity score system in which an immunoreactivity score is generated by incorporating both the percentage of positive tumor cells (0 to 100) and the intensity of staining (0 to 3). Briefly, slides were scored by the cross-product (H score) of the percentage of tumor cells staining at each of three staining intensities (*24*) and a final H score (numbers range from 0 to 300) was determined for each case. GLUT4 expression was also correlated with response to imatinib.

KIT and *PDGFRA* mutations were determined using polymerase chain reaction analysis of genomic DNA obtained from tumor biopsies (n= 31) followed by direct sequencing as previously reported (*20*)



Supplemental Figure 1. Enrollment summary.



Supplemental Figure 2. IHC analysis of **A)** CD117 and **B)** GLUT 1 on an untreated GIST biopsy, GLUT 1 staining on **C)** a lung carcinoma and **D)** an invasive ductal carcinoma breast sample, and **E)** CD117 and **F)** GLUT 4 on an untreated GIST biopsy. The majority of pre-treatment GIST samples had high expression of GLUT 4 and low to absent GLUT 1 expressions as shown by IHC.

GLUT 1 IHC showed moderate to high expression in a number of carcinoma samples tested as expected.