Supplemental online content for:

A Polycythemia Vera JAK2 Mutation Masquerading as a Duodenal Cancer Mutation

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eAppendix 1: Supplemental Methods
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Sample Collection, DNA Extraction, and Sequencing
Tumor and adjacent normal tissue samples were obtained as formalin-fixed paraffin-embedded slides prepared for DNA analysis as previously described.\(^1\) Tumor tissue was sent for commercial next-generation sequencing (NGS) testing (Foundation Medicine, Inc., Cambridge, MA) using a cancer gene panel (FoundationOne). Confirmatory NGS of tumor and normal tissue was performed in the Johns Hopkins Molecular Pathology Laboratory using the Ion Torrent Ampliseq v2.0 cancer gene panel (ThermoFisher Scientific, Waltham, MA). Other sources of germline tissues (buccal swab, fingernail clippings) were obtained and DNA extracted per the manufacturer’s protocol (Qiagen, Hilden, Germany). These samples were used for Sanger capillary sequencing as described.\(^2\) Research protocols were approved by the Johns Hopkins Institutional Review Board. Primers used for polymerase chain reaction amplification were 5’-TTCAGGTGTTATGGGTCAAGC-3’ (forward) and 5’-TGAAAAGGCCAGTTATTCCAA-3’ (reverse), which amplified 698 base pair products that were gel-purified and subjected to Sanger sequencing for both sense and antisense strands using the 5’-AAAGGGACCAAAGCACATTG-3’ (forward) and 5’-TGTTTGGGCATTGTAACCTTC-3’ (reverse) primers, respectively.

References