

LETTER TO THE EDITOR

Absence of JAK2 V617F mutation in gastric cancers

JONG WOO LEE¹, YOUNG HWA SOUNG¹, SU YOUNG KIM¹, SUK WOO NAM¹,
WON SANG PARK¹, JUNG YOUNG LEE¹, NAM JIN YOO¹ & SUG HYUNG LEE¹

¹Department of Pathology, College of Medicine, The Catholic University of Korea, Seoul, Korea

To the Editor:

Protein tyrosine kinases, which regulate cell-signaling pathways mediating a number of processes in cell survival and growth, comprise a large fraction of the dominant oncogenes known to date. Janus kinase (JAK) proteins are intracellular kinases that provide important links between cytokine receptors and down stream effector proteins [1]. Recently, a missense somatic mutation in the pseudokinase domain of *JAK2* gene (*JAK2* V617F) has been reported in chronic myeloproliferative disorders. The mutation was observed in most polycythemia vera (up to 97%) as well as in about one third of both essential thrombocythemia and myeloid metaplasia with myelofibrosis [2,3]. Functionally, this mutation constitutively activates JAK2 signaling, suggesting its role in the neoplastic transformation [2,3]. Furthermore, a small molecule inhibitor of JAK2 led to inhibition of the JAK2 V617F-possessing leukemic cell proliferation *in vitro* [2], thus suggesting that the JAK2 mutant is a promising molecular target for cancer therapy. Because activation of JAK signaling frequently occurs in other malignancies as well including, gastric cancers [4], we investigated the occurrence of the JAK2 mutation in gastric cancers by a polymerase chain reaction (PCR)-single strand conformation polymorphism (SSCP) assay.

Methacarn-fixed tissues of 180 gastric adenocarcinomas were randomly selected for the study. All of the patients were Asians (Korean). The gastric carcinomas consisted of 79 diffuse-type, 63 intestinal-type and 38 mixed-type gastric adeno-

carcinomas by Lauren's classification, and 40 early and 140 advanced gastric carcinomas according to the depth of invasion. In 58 gastric adenocarcinomas we analyzed the mutation both in primary tumors and metastatic tumors in the regional lymph nodes, and in the remaining 122 tumors we analyzed the mutation in primary tumors. The TNM stages of the gastric carcinomas were 81 stage I, 44 stage II, 40 stage III and 15 stage IV. Malignant cells and normal cells from the same patients were selectively procured from hematoxylin and eosin-stained slides using a 30G1/2 hypodermic needle (Becton Dickinson, Franklin Lakes, NJ) affixed to a micro-manipulator by microdissection.

To date, all of the JAK2 mutations reported have been 1849G > T in the exon 12 which would result in an amino acid substitution (V617F) within the JH2 pseudokinase domain [2,3]. Thus, we analyzed the exon 12 of JAK2 gene by PCR-SSCP. Genomic DNA each from tumor cells and normal cells were amplified by PCR with one primer pair (5'-tgga-caacagctcaaacaac -3' and 5'-ctgagaaaggcattagaaaag -3') covering the exon 12. Radioisotope was incorporated into the PCR products for detection by SSCP autoradiogram.

However, the SSCP from the 180 tumors did not reveal any aberrantly migrating band compared to the wild-type bands from the normal tissues. To confirm the SSCP results, we repeated the experiments twice, including tissue microdissection, PCR, SSCP and direct DNA sequencing analysis to ensure the specificity of the results, and found that the data were consistent. By comparison, we found the JAK2

V617F mutation in 65% (11/17) of the bone marrows of the polycythemia vera patients in agreement with the previous study [2,3], demonstrating that our PCR-SSCP methods could detect the JAK2 V617F mutation.

Discovery of the JAK2 mutation in chronic myeloproliferative disorders raised several critical questions. One of them was as to whether solid tumors harbor the same mutation, because JAK signaling is constitutively activated in many of solid tumors [4]. In the present study, we have analyzed the JAK2 V617F mutation in gastric cancers, one of the leading causes of cancer deaths worldwide, and found no JAK2 V617F mutation in the samples. This observation indicated that the JAK2 gene mutation rarely occurs in gastric cancers and suggests that the activation of JAK signaling previously observed in gastric cancer may not be related to the JAK2 mutation. Along with this study, we also analyzed the JAK2 mutation in other common malignancies including, colon cancers, breast cancers and lung cancers, but found no JAK2 V617F mutation (unpublished data), suggesting that the JAK2 mutation is limited in the hematopoietic neoplasia. Therapeutically, mutated tyrosine kinases have become rational targets for cancer treatments, and the most impressive examples of recent cancer therapies used the kinase inhibitors such as Imanitib

(Gleevec), Trastuzumab (Herceptin) and Gefitinib (Iressa) [5]. However, the present data showing no JAK2 mutation in gastric cancers strongly suggested the low possibility of targeting mutated JAK2 in these cancers.

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