JAK2V617F, an acquired mutation of JAK2, is present in a majority of patients with polycythemia vera and to a lesser extent among patients with the other myeloproliferative disorders. We analyzed the effect of JAK2V617F on the expression of polycythemia rubra vera 1 (PRV-1), using an in vitro model. Compared to wild-type JAK2, the presence of JAK2V617F increased both PRV-1 protein and mRNA levels in murine myeloid cells. A JAK2 inhibitor eliminated the V617F-induced increase in PRV-1 expression.

Figure 1. JAK2 mutation and expression of PRV-1. A. Expression of PRV-1 on FDCP-JAK2V617F, FDCP-JAK2WT, and FDCP-sham cells was compared by flow cytometry. The results of one of the experiments are shown as histograms. Binding of a phycoerythrin-conjugated secondary antibody served as the negative control. B. Results of flow cytometry experiments are summarized as a bar graph. MFI (PRV-1)/MFI (JAK2) means a higher expression of PRV-1. A lower ΔΔCt means a higher expression of PRV-1 mRNA.

The effect of the JAK2 V617F mutation on PRV-1 expression

JAK2V617F, an acquired mutation of JAK2, is present in a majority of patients with polycythemia vera and to a lesser extent among patients with the other myeloproliferative disorders. We analyzed the effect of JAK2V617F on the expression of polycythemia rubra vera 1 (PRV-1), using an in vitro model. Compared to wild-type JAK2, the presence of JAK2V617F increased both PRV-1 protein and mRNA levels in murine myeloid cells. A JAK2 inhibitor eliminated the V617F-induced increase in PRV-1 expression.

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sequence and its intron-exon boundaries were determined by using the reported murine PRV-1 mRNA (NM_026862) and the NCBI Map Viewer website. Total RNA was extracted from the transduced FDCP cells, reverse transcribed to double-stranded DNA using a cell to cDNA kit (Ambion), and used as a template in the real-time PCR. We used the 18S RNA as an internal standard and normalized all the real-time PCR results to that of the FDCP-Sham cells. FDCP-JAK2V617F cells had significantly higher amounts of PRV-1 mRNA than had FDCP-JAK2WT cells (Figure 1C; ΔΔCt: -9.8±0.3 and -1.7±0.8, respectively; p<0.05). To investigate whether the JAK2 mutation is the cause of PRV-1 overexpression, we studied the effect of a JAK2-specific tyrosine kinase inhibitor on PRV-1 expression. This inhibitor, 1,2,3,4,5,6-hexabromocyclohexane (HBCH), was obtained from the Drug Synthesis and Chemistry Branch, developmental Therapeutics Program, Division of Cancer Treatment and Diagnosis, National Cancer Institute. About 10^6 FDCP...
cells in culture media supplemented with the necessary growth factors were incubated in the presence of different concentrations of HBCH (0, 1, 5, 20, and 40 \(\mu\)M) for 24 hours. HBCH (particularly at concentrations ranging from 1 to 5 \(\mu\)M) decreased PRV-1 expression on FDCP-JAK2\textsuperscript{V617F} cells (Figure 2), but did not significantly alter the expression of PRV-1 in FDCP-JAK2\textsuperscript{WT} cells. Our results show that mutation in JAK2 could be responsible for an increase in the PRV-1 expression in MPD. One intriguing question is whether PRV-1 overexpression is only a phenotypic change or whether it has any functional consequence. We have previously shown that overexpression of PRV-1 in a heterologous cell line can promote proliferation in the absence of growth factors.\textsuperscript{7} Considering our observations, we put forward the following hypothesis: in MPD, JAK2\textsuperscript{2\(\mu\)M} is the cause of PRV-1 overexpression and the latter increases cell proliferation. This hypothesis should be tested by studying the effect of selective inhibition of PRV-1 (in the presence of JAK2\textsuperscript{2\(\mu\)M}) on cell proliferation.

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References