Erythroid lineage-restricted expression of Jak2V617F is sufficient to induce a myeloproliferative disease in mice

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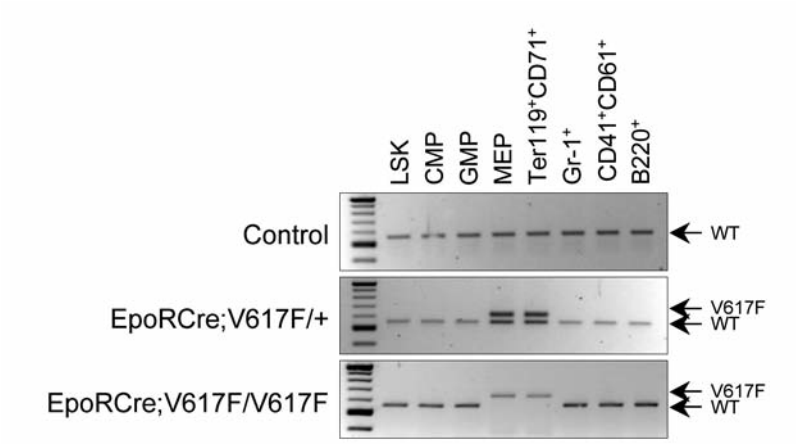
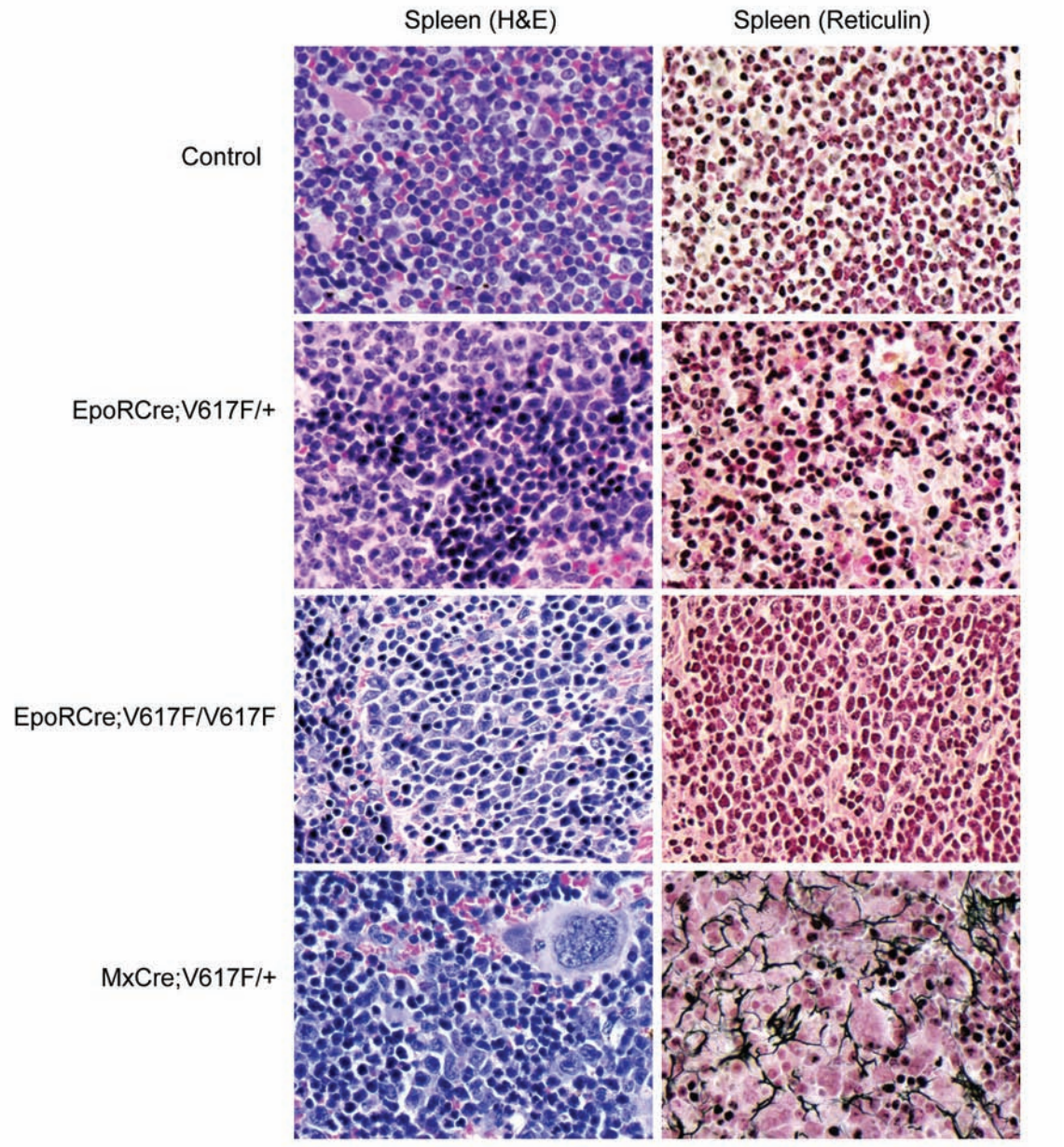
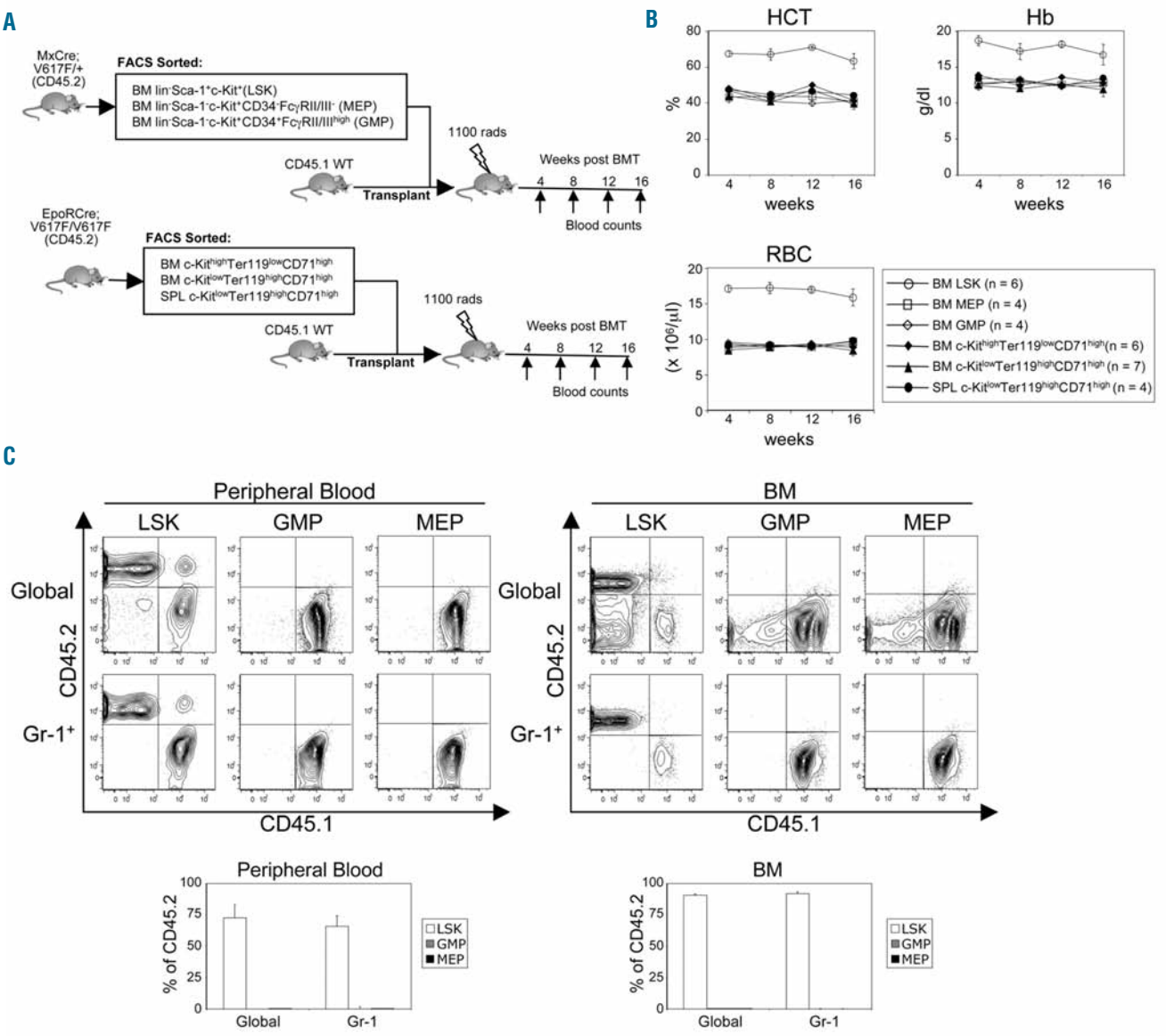


Online Supplementary Figure S1. EpoRCre induces deletion of the loxP flanked cassette in MEP and erythroid progenitors only. Deletion of the loxP flanked PGK-Neo-Stop cassette was analyzed in FACS sorted LSK, CMP, GMP, MEP, Ter119$^+$CD71$^+$, Gr-1$^+$, CD41$^+$CD61$^+$ and B220$^+$ cells from control, EpoRCre;V617F/+, and EpoRCre;V617F/V617F mice BM by PCR. The presence/expression of the wild-type (WT) and Jak2V617F (V617F) alleles are shown. Note that EpoR-Cre mediated recombination/deletion of the PGK-Neo-Stop cassette with subsequent expression of Jak2V617F was observed specifically in MEP and erythroid (Ter119$^+$CD71$^+$) populations.



Online Supplementary Figure S2. Histopathological analyses. Hematoxylin and eosin (H&E) staining shows expansion of erythroid precursors in the spleens of EpoRCre;V617F/+ and EpoRCre;V617F/V617F mice compared with controls (left panels; 500X). Reticulin staining demonstrates the absence of fibrosis in the spleens of these animals (right panels; 500X). H&E staining of spleen sections from induced MxCre;V617F/+ mice display increased numbers of megakaryocytes, erythrocytes and myeloid cells (left bottom panel; 500X) and reticulin staining shows the presence of fibrosis in the spleens of MxCre;V617F/+ mice (right bottom panel; 500X).



Online Supplementary Figure S3. Jak2V617F-expressing committed progenitors (GMP, MEP) or erythroid progenitors could not transfer the disease into secondary recipients. (A) Experimental design for transplantation of LSK, GMP, MEP, or erythroid progenitors into secondary recipients. Sorted LSK (2 x 10^4), MEP (2 x 10^4), or GMP (2 x 10^4) from the MxCre;V617F/+ mice or erythroid progenitors (10^5) from the EpoRCre;V617F/V617F mice were mixed with CD45.1$^+$ wild-type BM cells (10^5) and transplanted into lethally irradiated recipient mice (CD45.1$^+$). (B) Peripheral blood hematocrit, hemoglobin and RBC counts at 4, 8, 12, and 16 weeks after transplantation are shown. (C) Representative contour plots on the ratio of CD45.2$^+$ to CD45.1$^+$ in all hematopoietic cells (global) and in Gr-1$^+$ population in the peripheral blood and BM are shown (upper panels). Histograms show percentage of CD45.2$^+$ cells (calculated as CD45.2$^+$/CD45.1$^+$ plus CD45.2$^+$) in the peripheral blood leukocytes and BM of recipient animals (lower panels). Results are shown as mean ± SEM. Note that Jak2V617F confers self-renewal capacity only to LSK but not GMP, MEP or erythroid progenitors.