Deletion of miR-451 curbs JAK2(V617F)-induced erythrocytosis in polycythemia vera by oxidative stress-mediated erythroblast apoptosis and hemolysis

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Supplemental Fig. S1. Representative flow cytometric plots of bone marrow and spleen EryA, EryB and EryC cells. Percentages of each population are shown.



Supplemental Fig. S2. Quantification of erythroblast subsets based on morphology under May-Grünwald-Giemsa staining. Proerythroblast and basophilic erythroblast (Pro/Baso), polychromatic erythroblast (Poly), and orthochromatic erythroblast (Ortho) from WT-J2VF and KO-J2VF bone marrow and spleen were quantified.



Supplemental Fig. S3. Quantitative analyses of ROS in erythroblast subsets. (A) ROS levels in GFP+ EryA, EryB and EryC cells from WT-J2WT, WT-J2VF, KO-J2WT and KO-J2VF mice. (B) Quantification of ROS-positive percentages in EryC cells. Histogram overlays are shown with cell numbers normalized to mode (top) or cell numbers (bottom) y-axis. ***p<0.001.



Supplemental Fig. S4. Quantitative analyses of apoptosis in erythroblast subsets. Annexin V+7AAD- apoptotic GFP+ EryA, EryB and EryC cells were quantified in bone marrow and spleen. *p<0.05.



Supplemental Fig. S5. Fewer free thiols are detected in KO-J2VF RBCs. RBCs were stained with 2.5μ M ThiolTracker Violet (ThermoFisher) for 30 min in a standard tissue culture incubator (37°C, 5% CO2). Median fluorescence intensity (MFI) in Kusabira Orange negative cells was analyzed by flow cytometry. ***p<0.001.