SUPPLEMENTARY INFORMATION

SUPPLEMENTARY NOTE 1: Reticulocyte maturation, measured by the intensity of fluorescence (56), was normal in *Gas6*$^{-/-}$ mice. The proportion of immature reticulocytes with a high fluorescence versus mature reticulocytes with an intermediate or low fluorescence were, respectively, 12 ± 0.4% and 89 ± 1.9% in WT mice versus 11 ± 1.1% and 88 ± 0.6% in *Gas6*$^{-/-}$ mice (n = 3, P = NS).

SUPPLEMENTARY NOTE 2: RBCs were biotinylated in vivo. At 5 and 8 days after injection of the 34-3C-antibody, 62 ± 2% (day 5) and 39 ± 1% (day 8) biotinylated rbcs were detected in WT mice versus 66 ± 2% (day 5) and 43 ± 2% (day 8) in *Gas6*$^{-/-}$ mice, respectively (n = 6, P = NS).

SUPPLEMENTARY NOTE 3: Since iron reserves are necessary for a sufficient erythropoietic response and iron depletion is a common cause of resistance to Epo, we assessed the mice for iron stores. Perl's staining of BM cytospins revealed adequate iron stores in both WT and *Gas6*$^{-/-}$ mice (data not shown). Serum iron levels were also within the normal range (data not shown). In addition, BM histology revealed normal stainable iron stores in both WT and *Gas6*$^{-/-}$ mice (not shown). Thus, the impaired erythropoietic response in anemic *Gas6*$^{-/-}$ mice, in spite of elevated Epo levels (see Supplementary Note 4), was not attributable to iron deficiency.

SUPPLEMENTARY NOTE 4: Since maturation and proliferation of erythroid progenitors, as well as the generation of early erythroblasts are largely dependent on Epo (2), we considered whether the impaired erythropoietic response of *Gas6*$^{-/-}$ mice might be due to insufficient Epo production. In baseline conditions, serum Epo protein levels were undetectable in both genotypes (< 20 mIU/ml). Remarkably, after PHZ-induced hemolysis, serum Epo levels were higher in *Gas6*$^{-/-}$ than WT mice (mIU/ml: 53 ± 12 and 25 ± 0.5 in WT mice versus 440 ± 120 and 165 ± 50 in *Gas6*$^{-/-}$ mice at day 3 and 6 after PHZ, respectively; n = 4-7, P < 0.05). Thus, *Gas6*$^{-/-}$ mice displayed higher erythropoietin levels at day 3 and 6 after acute hemolysis than WT mice because they were more anemic at these time points (Figure 2*a*).

SUPPLEMENTARY NOTE 5: RBCs were biotinylated in vivo in WT and *Gas6*$^{-/-}$ mice. rGas6 was administered from day 0 to day 8 (2 μg/day, i.p.). On day 0, autoiimmune hemolytic anemia was induced by intraperitoneal injection of 200 μg purified anti-RBC 34-3C IgG2a (29). At 5 and 8 days after administration of rGas6 and anti-RBC antibodies, respectively, the percentage of biotinylated RBCs was 53 ± 3% and 39 ± 2% in WT mice versus 55 ± 7% and 37 ± 2% in *Gas6*$^{-/-}$ mice (n = 6, P = NS), indicating that the clearance rate of biotinylated RBCs during acute hemolytic anemia was comparable in *Gas6*$^{-/-}$ than WT mice in presence of rGas6 (see also Supplementary note 2).

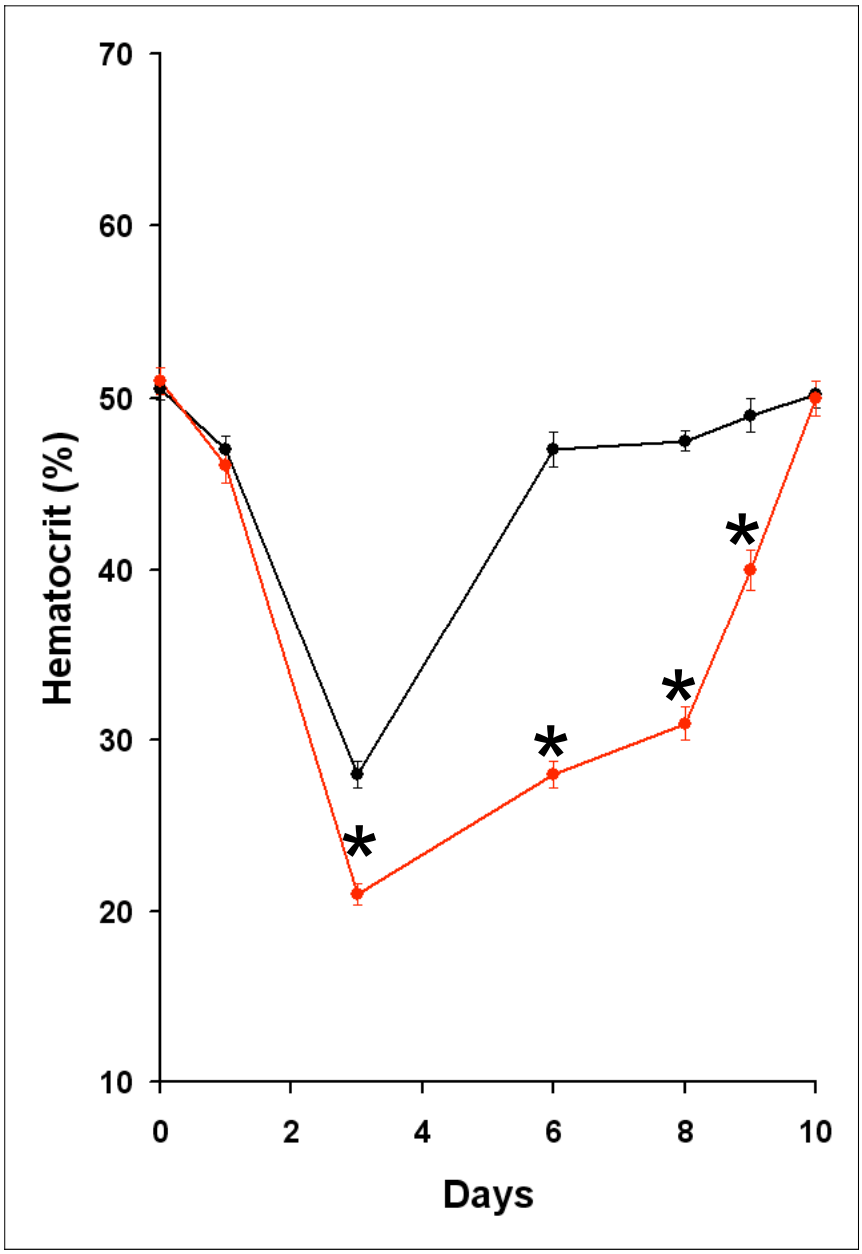
56. Watanabe, K., et al. 1994. Reticulocyte maturity as an indicator for estimating qualitative abnormality of erythropoiesis. J. Clin. Pathol. 47:736–739.

SUPPLEMENTARY FIGURES

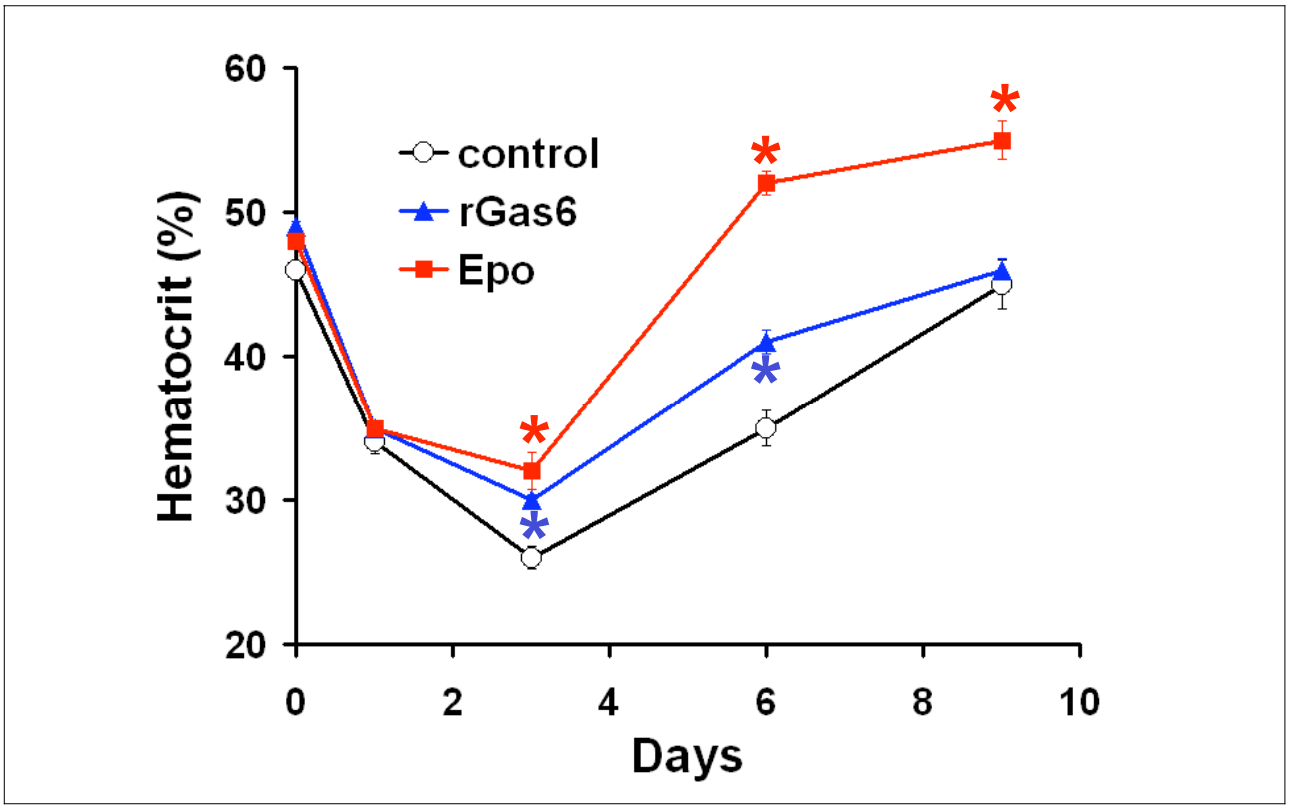
SUPLEMENTARY FIGURE 1: Impaired erythropoiesis in *Gas6*$^{-/-}$ mice (red lines) as compared to WT mice (black lines) on a 100% C57BL/6 background in response to phenylhydrazine induced anemia (injection on day 0 and 1).

SUPPLEMENTARY FIGURE 2: Therapeutic potential of recombinant Gas6 in acute anemia induced by blood loss.

After bleeding (500 μl on day 0 and 500 μl on day 1), WT mice were treated with saline (control), recombinant human Gas6 (rGas6, 2 μg daily intraperitoneally) or recombinant human erythropoietin (Epo, 10 IU every second day intraperitoneally). The erythropoietic response was monitored by determining the hematocrit levels. Hematocrit levels are expressed as mean ± SEM (n = 6 mice) in all panels. *, $P < 0.001$.



Supplementary Fig. 1



Supplementary Fig. 2