Supplemental figure 1

A

B

$r = 0.62$
$p < 0.01$

$r = -0.68$
$p < 0.001$
Supplemental Figure 2

A. Iron concentration at e17.5 (ppm)

B. Relative hamp expression at e17.5

C. FPN transactivation in fetal livers

D. Normalised intensity

E. FPN transactivation in fetal livers

F. Normalised intensity

G. Serum iron concentration at e17.5 (μM)
Supplemental figure 3

A

- hamp^fl/fl
- hamp^fl/fl, Alb.Cre+

p < 0.05

B

- Liver iron concentration at e17.5 (ppm)

200ppm 5ppm

p < 0.001

p < 0.0001

n.s.
Supplemental figure 4

A. Relative hamp expression in liver at e17.5

B. Liver iron concentration at e17.5 (ppm)

C. Relative fpr expression in liver at e17.5

D. Relative expression of tfr1 and dmt1 in liver at e17.5

E. Relative expression of fprC326Yfl/fl and fprC326Yfl/fl, Alb.cre+

F. Relative expression of hamp fl/fl and hamp fl/fl, Alb.cre+

G. Relative tfr1 expression at e17.5

H. Relative dmt1 expression at e17.5
### Supplemental Figure 5

**A**

- $fpn^{wt/wt}$
- $fpn^{wt/C326Y}$

Normalised intensity

**B**

- $fpn^{wt/wt}$
- $fpn^{wt/C326Y}$

Normalised intensity

**C**

- $fpn^{C326Y^{fl/fl}}$

Normalised intensity

**D**

- $fpn^{C326Y^{fl/fl}, Alb.cre+}$

Normalised intensity

**E**

- $hamp^{fl/fl}$

Normalised intensity

**F**

- $hamp^{fl/fl, Alb.cre+}$

Normalised intensity

- *p < 0.05
- n.s.
LEGENDS FOR SUPPLEMENTAL FIGURES

Supplemental Figure 1: A. Correlation between hemoglobin and the percentage of R5 cells in fetal livers at e17.5 (n=21). B. Correlation between hemoglobin and the percentage of R3 cells in fetal livers at e17.5 (n=22). P values are calculated using Student’s t test. r is Pearson’s correlation coefficient.

Supplemental Figure 2: fpn<sup>wt/wt</sup> animals were derived from fpn<sup>wt/wt</sup> mothers that were either fed a control diet containing 200ppm iron or an iron-loaded diet containing 5000ppm iron throughout pregnancy. A. Iron concentration in the maternal liver (n=5 and 3, respectively), fetal liver (n=3, 4) and placenta (n=3, 7) at e17.5. B. Relative hamp mRNA expression in the maternal liver (n=5, 3), fetal liver (n=3, 4) and placenta (n=4, 7) at e17.5. C-D. Western blot for FPN in the fetal livers at e17.5 (n=3, 3). E-F. Western blot for FPN in the placentae at e17.5 (n=3, 3). G. Iron concentration in fetal serum at e17.5 (n=11, 7). Values are shown as mean±S.E.M. P values are calculated using Student’s t test.

Supplemental Figure 3: A. Relative hamp mRNA expression in livers of hamp<sup>fl/fl</sup> animals and hamp<sup>fl/fl</sup>, Alb.Cre<sup>+</sup> littermates at e17.5 (n=4, 5 respectively). B. Liver iron concentration in hamp<sup>fl/fl</sup> animals and hamp<sup>fl/fl</sup>, Alb.Cre<sup>+</sup> littermates at e17.5. Animals were derived from hamp<sup>fl/fl</sup> mothers that were either fed a control diet containing 200ppm iron (n=8, 8) or an iron-deficient diet containing 5ppm iron (n=5, 4), from weaning and throughout pregnancy. Values are shown as mean±S.E.M. P values are calculated using Student’s t test.

Supplemental Figure 4: (A-C) fpn<sup>wt/wt</sup> animals were derived from fpn<sup>wt/wt</sup> mothers that were either fed a control diet containing 200ppm iron or an iron-deficient diet containing 5ppm iron, from weaning and throughout pregnancy. A. Relative hamp mRNA expression in livers at e17.5 (n=3, 3 respectively). B. Iron concentration in livers at e17.5 (n=8, 5). C. Relative fpn mRNA expression in livers at e17.5 (n=4, 3). D. Relative expression of tfr1 and dmt1 mRNA transcripts in livers of fpn<sup>wt/wt</sup> animals and fpn<sup>wt/C326Y</sup> littermates at e17.5 (n=6, 7, respectively). E. Relative expression of tfr1 and dmt1 mRNA transcripts in livers of fpn<sup>C326Y/fl</sup> animals and fpn<sup>C326Y/fl</sup>, Alb.Cre<sup>+</sup> littermates at e17.5 (n=3, 5 respectively). F. Relative expression of tfr1 and dmt1 mRNA transcripts in livers of hamp<sup>fl/fl</sup> animals and hamp<sup>fl/fl</sup>, Alb.Cre<sup>+</sup> littermates at e17.5 (n=4, 4 respectively). G-H. fpn<sup>wt/wt</sup> animals were derived from fpn<sup>wt/wt</sup> mothers that were either fed a control diet containing 200ppm iron or an iron-deficient diet containing 5ppm iron, from weaning and throughout pregnancy. Relative expression of tfr1 and dmt1 mRNA transcripts in livers (n=4, 3 respectively) and placentae (n=5, 4). Values are shown as mean±S.E.M. P values are calculated using Student’s t test.

Supplemental Figure 5: Normalised intensities for FPN Western blots. A. Placentae of fpn<sup>wt/wt</sup> animals and fpn<sup>wt/C326Y</sup> littermates. B. Livers of fpn<sup>wt/wt</sup> animals and fpn<sup>wt/C326Y</sup> littermates. C. Placentae of fpn<sup>C326Y/fl</sup> animals and fpn<sup>C326Y/fl</sup>, Alb.Cre<sup>+</sup> littermates. D. Livers of fpn<sup>C326Y/fl</sup> animals and fpn<sup>C326Y/fl</sup>, Alb.Cre<sup>+</sup> littermates. E. Placentae of hamp<sup>fl/fl</sup> animals and hamp<sup>fl/fl</sup>, Alb.Cre<sup>+</sup> littermates. F. Livers of hamp<sup>fl/fl</sup> animals and hamp<sup>fl/fl</sup>, Alb.Cre<sup>+</sup> littermates. N=3 mice per group. Values are shown as mean±S.E.M. P values are calculated using Student’s t test.