Supplemental Figure 1. Hepatic and splenic CCS protein levels in CuA and CuD rats. Western blot analysis of hepatic CCS in postweaning (A) and perinatal (B) copper deficiency. Western blot analysis of splenic CCS in postweaning (C) and perinatal (D) copper deficiency. To indicate protein loading, the blot was stripped and reprobed for tubulin. Values below Western blots indicate relative intensities of CCS-immunoreactive bands. Values represent mean ± SEM, n = 3-4. *Different from respective CuA group, P < 0.05.
Supplemental Figure 2. Hepatic TfR2 protein and BMP6 mRNA levels in rats under various iron conditions. Weanling male Sprague-Dawley rats were fed modified AIN-93G purified rodent diets analyzed to contain 10 ppm Fe (FeD), 50 ppm Fe (FeA) or 18916 ppm Fe (FeO) for 3 wk. (A) Western blot analysis of crude membrane proteins from the livers of 6-wk-old FeA or FeD rats was used to demonstrate the decrease in TfR2 protein levels that occurs when plasma iron levels are low. To indicate protein loading, the blot was stripped and reprobed for SR-B1. Plasma iron levels (µmol/L) are indicated below each lane. (B) Q-RT-PCR analysis of BMP6 mRNA levels in livers of 6-wk-old FeO, FeA and FeD rats were used to demonstrate the modulation in BMP6 mRNA levels that occurs with varying iron status. Group means were compared by 1-way ANOVA followed by Tukey’s multiple comparison test. Tissue non-heme iron levels (µmol/g) are as indicated below each bar. Values represent mean ± SEM, n=6/group. Means without a common letter differ, P < 0.05.