Supplementary Figure 1. **HNF4α re-expression is confined to only hepatocytes in rats with terminal decompensated liver function and cirrhosis.** Cirrhotic rats with severe liver failure that persisted four weeks after the last dose of CCL4 were given a recombinant AAV expressing HNF4α and GFP. Enlarged images of fluorescence staining are shown: GFP (green) for virus transduction, EpCAM (red) for bile duct cells, α–SMA (red) for hepatic stellate cells, and HNF4α (red), for hepatocytes. Dapi (blue) was used as counterstaining, magnification, X400. **Immunofluorescence was performed on specimens from one animal per group and is representative of four images per biologic sample.**

Supplementary Figure 2. **RNA-Seq analysis showing phenotypic restoration of decompensated cirrhosis by AAV-HNF4α treatment.** RNA-seq was carried out on intact liver specimens involving 3 experimental conditions, untreated normal liver (blue), decompensated cirrhosis 24-26 weeks (green), and decompensated cirrhosis 14 weeks after treatment with AAV-HNF4α (red). Sequence reads were aligned against the rat genome (rn5). Studies were done on 2 replicates. Data are presented as the mean and standard deviation normalized to the value from normal liver. RNA-seq confirmed expression changes (Figure 5) in genes encoding liver-enriched transcription factors, metabolism proteins, and transport proteins.

Supplementary figure 3. **HNF4α re-expression in livers from functionally decompensated livers with terminal cirrhosis shows a decrease in hepatocyte apoptosis.** (A) TUNEL staining, a marker for apoptosis, and (B) graph showing quantification of TUNEL staining in normal, functionally compensated, un-treated decompensated cirrhotic livers, and decompensated cirrhotic livers 2 weeks and 14 weeks after HNF4α re-expression in vivo. Magnification, X200. **Immunohistochemistry was performed on specimens from one animal per biologic group and is representative of four images per biologic sample.** Each value represents the mean ± SD. Statistical analyses were performed using the Tukey-Kramer multiple comparisons procedure and two-tailed Student's t-test.
Supplementary Figure 4. Hepatocytes from rats with terminal decompensated liver function following HNF4α re-expression show limited repopulation ability.

Hepatocytes from normal, functionally compensated cirrhotic, decompensated cirrhotic, and decompensated cirrhotic livers fourteen weeks after AAV-HNF4α therapy were transplanted into the spleens of retrorsine-treated Nagase analbuminemic rats that underwent 70% partial hepatectomy at the time of transplantation. Immunofluorescence staining for albumin was performed on liver sections of transplanted Nagase analbuminemic rats. Small clusters of albumin-staining engrafted hepatocytes, which stain red, were present throughout the liver at 28 day after transplantation. Hepatocytes from normal and functionally compensated cirrhotic livers showed excellent engraftment and expansion following transplantation, whereas repopulation with hepatocytes from functionally decompensated rats was limited even following HNF4α re-expression. Albumin staining correlates with the albumin levels detected by ELISA in Fig 6. Magnification, X200. Transplants were performed on five animals per group. Each group was infused with hepatocytes isolated from one animal that underwent each of the various interventions. Immunofluorescence was performed on specimens from one animal per group and is representative of four images per biologic specimen.

Supplementary figure 5. Endogenous HNF4α expression is restored by treatment with AAV-HNF4α. (A) Specific quantitative qPCR analyses for endogenous and AAV-mediated HNF4α expression in normal, functionally decompensated cirrhotic livers, and decompensated cirrhotic livers fourteen weeks after in vivo HNF4α re-expression. Each value represents the mean ± SD. (B) RNA-seq, carried out on liver specimens from untreated normal livers (blue), decompensated livers with cirrhosis 24-26 weeks (green), and decompensated livers with cirrhosis 14 weeks after treatment with AAV-HNF4α (red). Sequence reads were aligned against the rat genome (rn5) or the AAV-HNF4α encoding plasmid. Studies were done on 2 replicates except for plasmid alignments, which were carried out on single specimens. Data are presented as the mean and standard deviation normalized to the value from normal liver, except for the viral analysis, which was normalized to the control level of HNF4a. Decreased HNF4a expression in cirrhotic rats was reversed by treatment with AAV-HNF4a. The treatment-induced increase in
HNF4a expression was due almost entirely to correction of native gene expression since AAV-HNF4-specific transcripts (the 5-untranslated region of HNF4a and the downstream region encoding GFP) were expressed at much lower levels (note change in scale).

**Supplementary figure 6. Effect of treatment with AAV-HNF4α on histology in rats with terminal decompensated liver function and cirrhosis.** (A) Photomicrographs of hematoxylin and eosin and Masson’s trichrome stained liver sections and (B) quantification of fibrosis in normal, functionally compensated, un-treated decompensated cirrhotic rat livers, decompensated cirrhotic rat livers two weeks after AAV-GFP therapy, and two and fourteen weeks after AAV-HNF4α therapy. For (B) each value represent mean ± SD. Statistical analysis was performed among four groups (decompensated cirrhotic hepatocytes with and without AAV-GFP or AAV-HNF4a transduction (after 2 weeks or 14 weeks) (**P<0.001). Histology was performed on specimens from two animals for all biologic groups except decompensated cirrhosis/AAV-HNF4α, which was performed on specimens from one animal. Studies are representative of four images per biologic sample. Statistical analysis was performed using the Tukey-Kramer multiple comparisons procedure and two-tailed Student's t-test. Each value represents the mean ± SD.
Supplementary Figure 2

The figure shows relative expression levels of various genes under different conditions:

- **HNF1α**, **C/EBPα**, **FOXA2**, **PPARα**
- **A1AT**, **CYP3A23/3a1**, **F7**, **OTC**
- **ApoA2**, **ApoC3**, **ApoE**, **TAT**
- **TDO2**, **TF**, **TTR**, **ALB**

Each gene is represented by three bars indicating relative expression levels in:
- Normal liver
- Decompensated cirrhosis
- Decompensated cirrhosis / AAV-HNF4α (14 weeks)