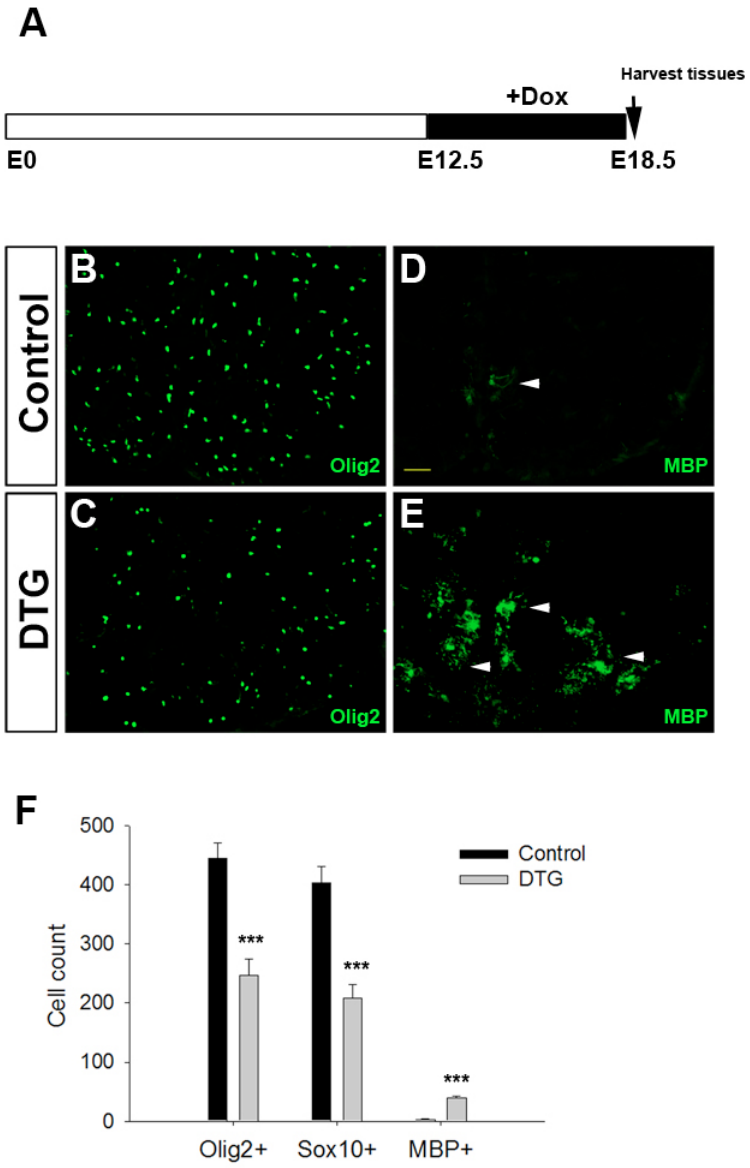

Fig. S1. Increased OL differentiation after long-term overexpression of *Nkx2.2* during embryonic period. (A) Schedule for Dox treatment. (B-E) Immunolabeling of anti-Olig2 (B-C), anti-MBP (D-E) after Dox exposure from E12.5 to E18.5. Many MBP+ cells, represented by white arrowheads, were found in the ventral spinal cord of DTG while only few in the control (D-E). (F) Quantification of Olig2+, Sox10+ or MBP+ cells per section in control and DTG mouse spinal cords (mean±s.e.m., n=3). ***: P<0.001. Scale bar: 50 μm.

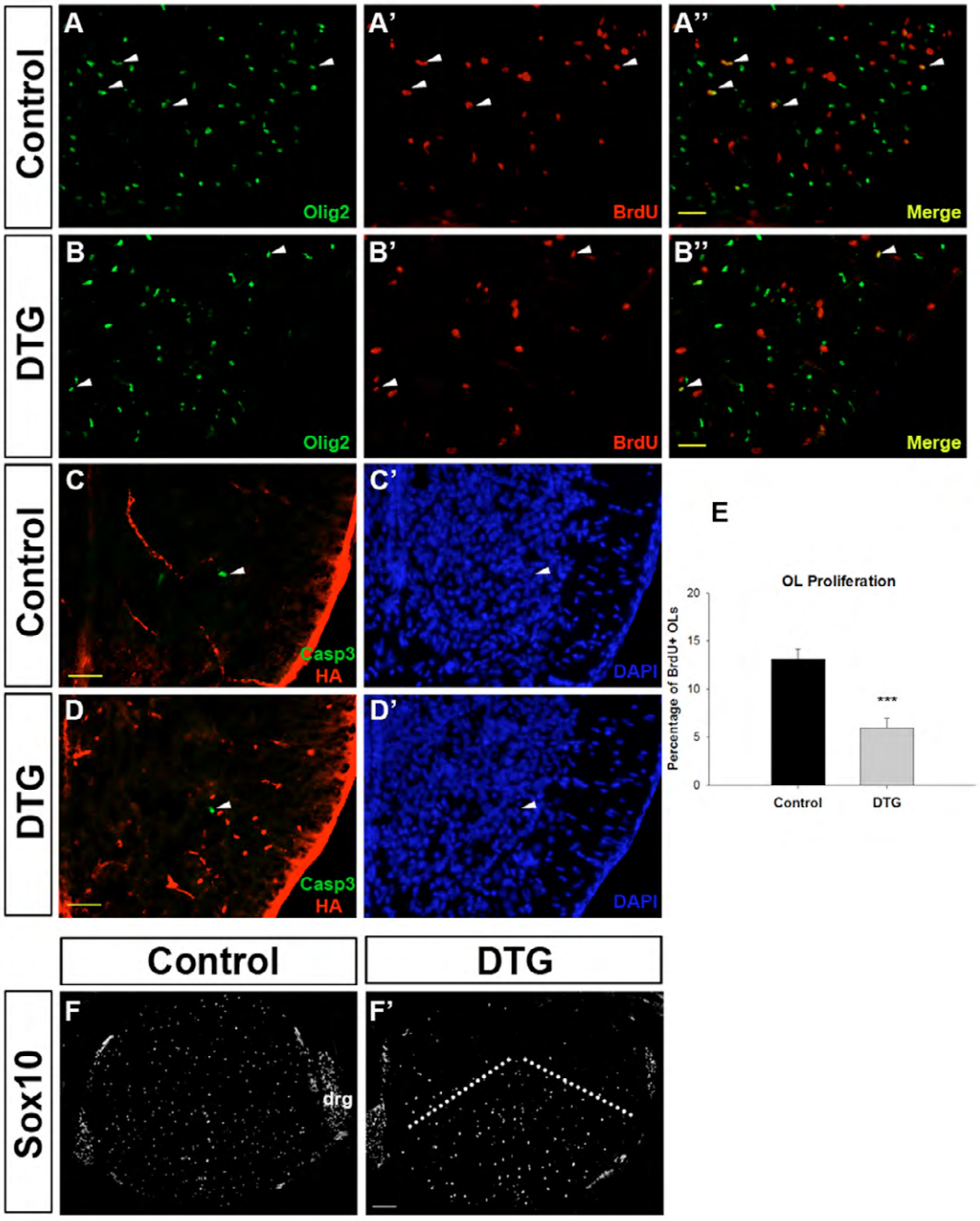

Fig. S2. Impaired OL proliferation and migration after induction of *Nkx2.2* in early OPCs. (A-D′) Double immunostaining of spinal cord sections from Dox-treated embryos (E12.5-E15.5) with anti-Olig2 and anti-BrdU (A-B”), anti-phospho Caspase-3 and anti-HA (C-D′). (E) Percentage of Olig2/BrdU double positive cells in Olig2+ cell population (mean±s.e.m., n=3). ***: P<0.001. (F-F′) The majority of Sox10+ OPCs in the DTG spinal cord were restricted to the ventral half. Scale bar: A-D′, 50 μm; F-F′, 100 μm.

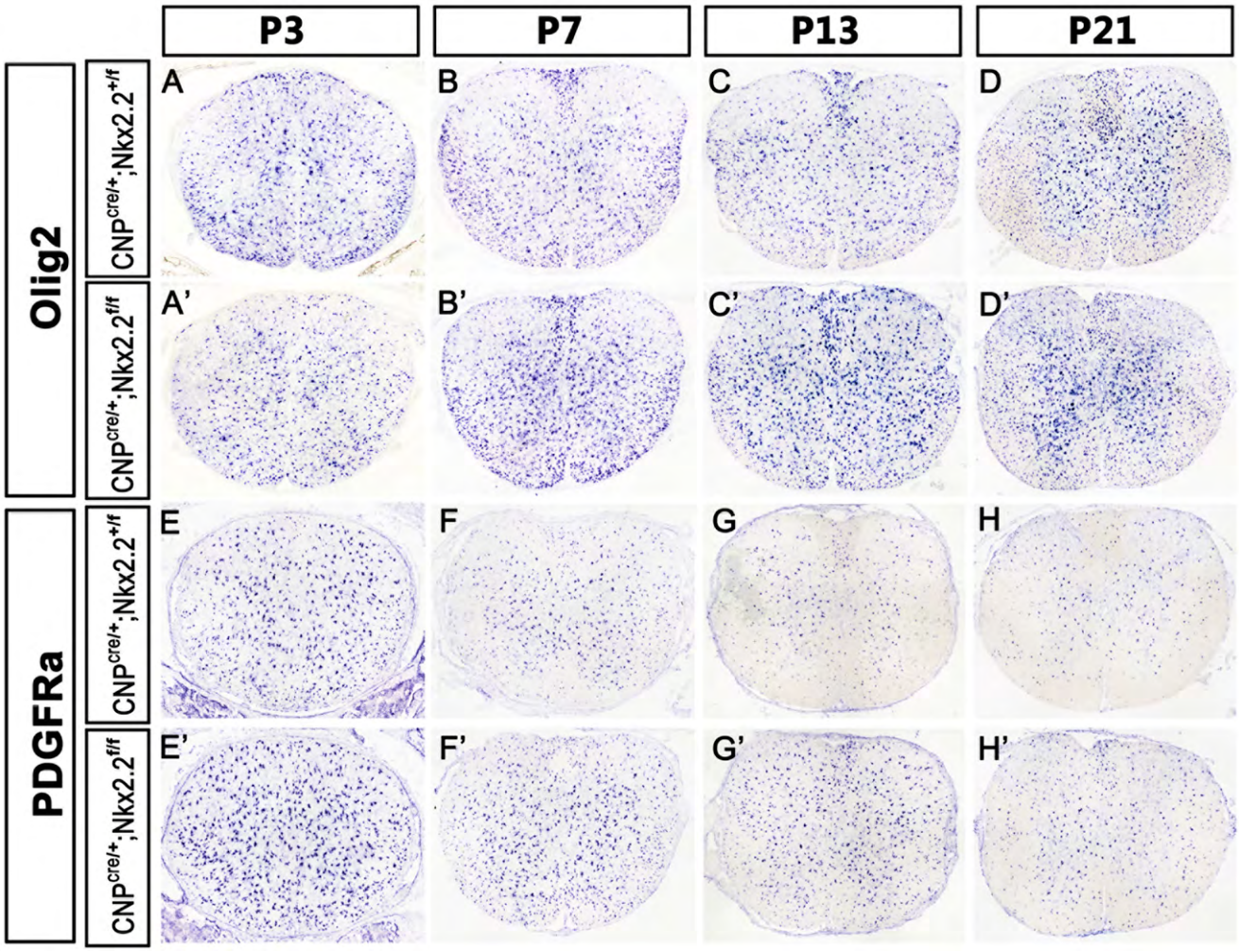

Fig. S3. Increased *PDGFRα* expression after conditional knockout of *Nkx2.2* in OLs. *In situ* hybridization of Olig2 (A-D′) and PDGFRα (E-H′) probes in spinal cords from $CNP^{+/cre}$;$Nkx2.2^{+/fl}$ and $CNP^{+/cre}$;$Nkx2.2^{fl/fl}$ from P3 to P21. There was an increase in the number of PDGFRα+ OPCs, but not the total number of Olig2+ OLs.

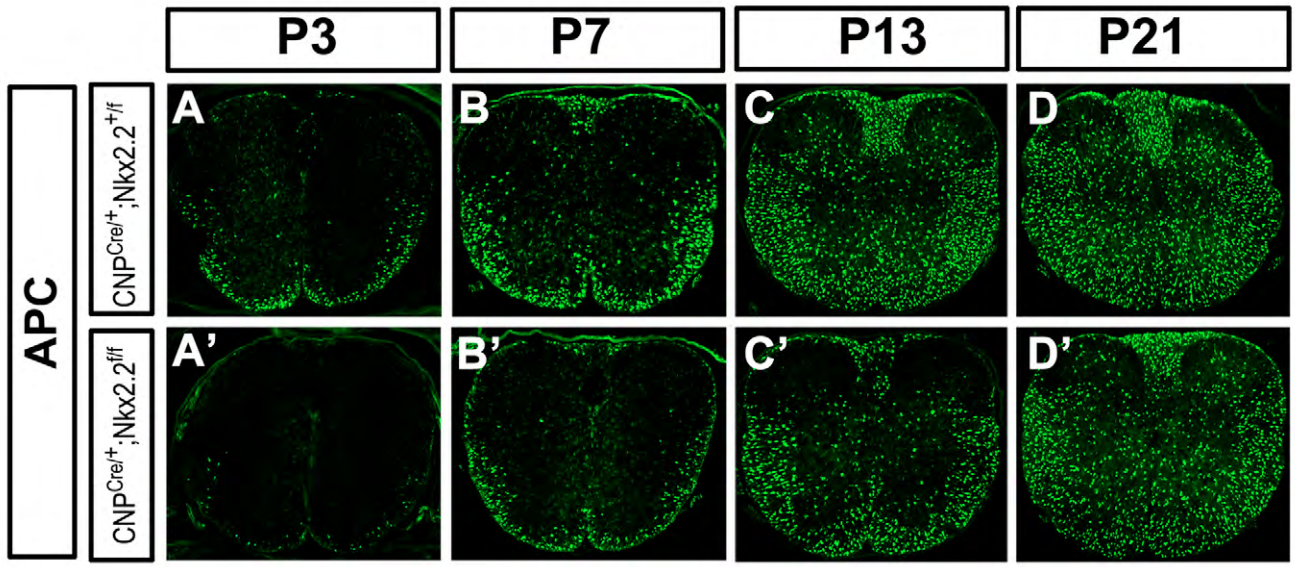

Fig. S4. Delayed expression of mature OL marker CC1 in *Nkx2.2* conditional knockout. Anti-CC1 immunostaining of spinal cord sections from $CNP^{+/cre}$;$Nkx2.2^{+/fl}$ and $CNP^{+/cre}$;$Nkx2.2^{fl/fl}$ at P3 (A-A′), P7 (B-B′), P13 (C-C′) and P21 (D-D′).

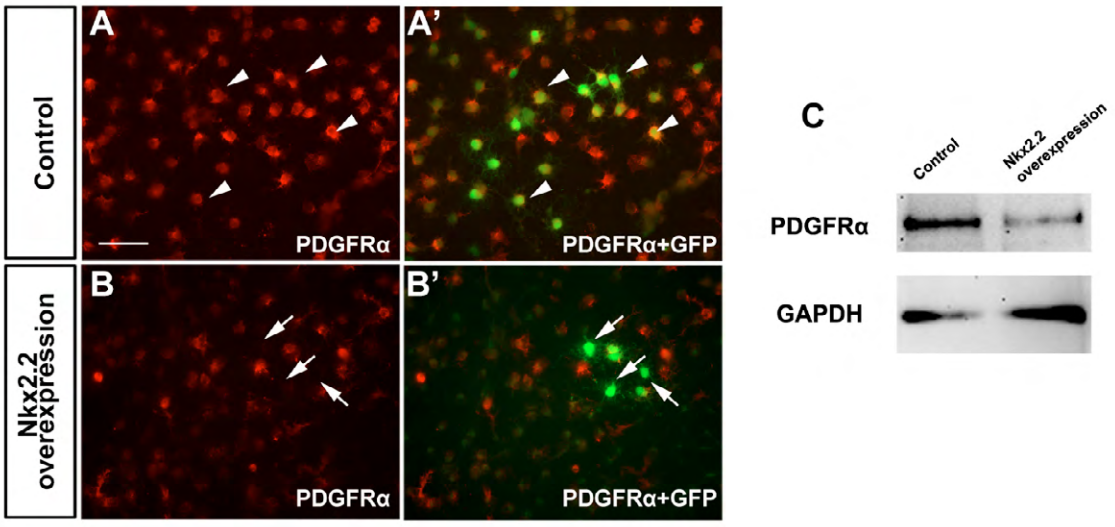

Fig. S5. Decreased *PDGFRα* expression after overexpressing Nkx2.2 in primary OPCs. (A)-B′) Immunostainig of *PDGFRα* in control (A-A′) and Nkx2.2-overexpression (B-B′) OPCs after the infection of lentivirus encodes GFP or Nkx2.2-GFP respectively. GFP expression did not affect *PDGFRα* expression, indicated by arrowheads in control (A-A′); while Nkx2.2-overexpression significantly suppressed *PDGFRα* expression, showed by arrow in (B-B′). (C) Detection of reduced PDGFRα in Nkx2.2-overexpressing OPCs by Western blot. GAPDH was loaded as internal control. Scale bar: 50 μm.

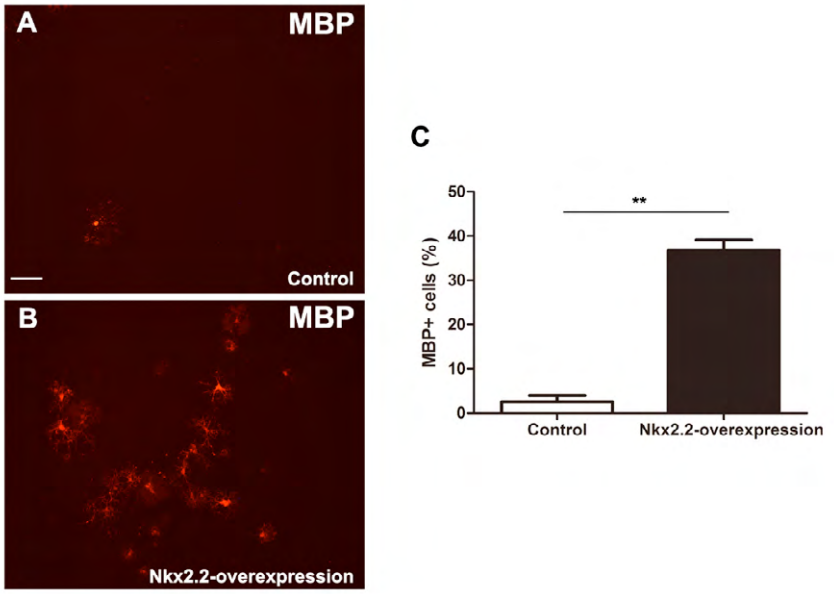

Fig. S6. Enhanced differentiation in primary OPCs overexpressing Nkx2.2. (A-B) Anti-MBP immunostaining of rat OPCs infected by control lentivirus (A) or lentivirus encoding Nkx2.2 (B). (C) Quantification of MBP+ cells in control and Nkx2.2-overexpression OPCs 6 days after lentivirus infection. **: $P<0.01$. Scale bar: 50 μm.