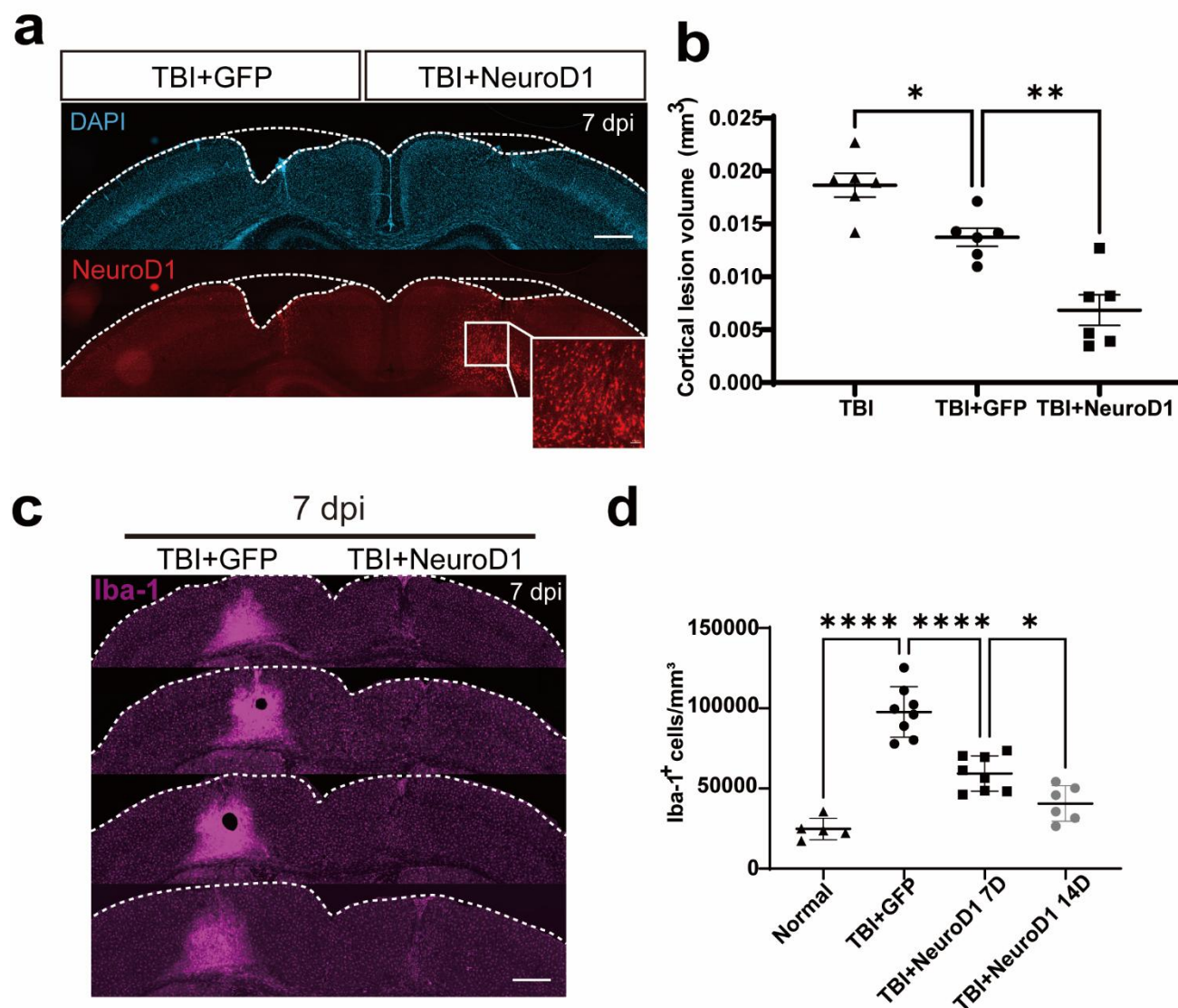


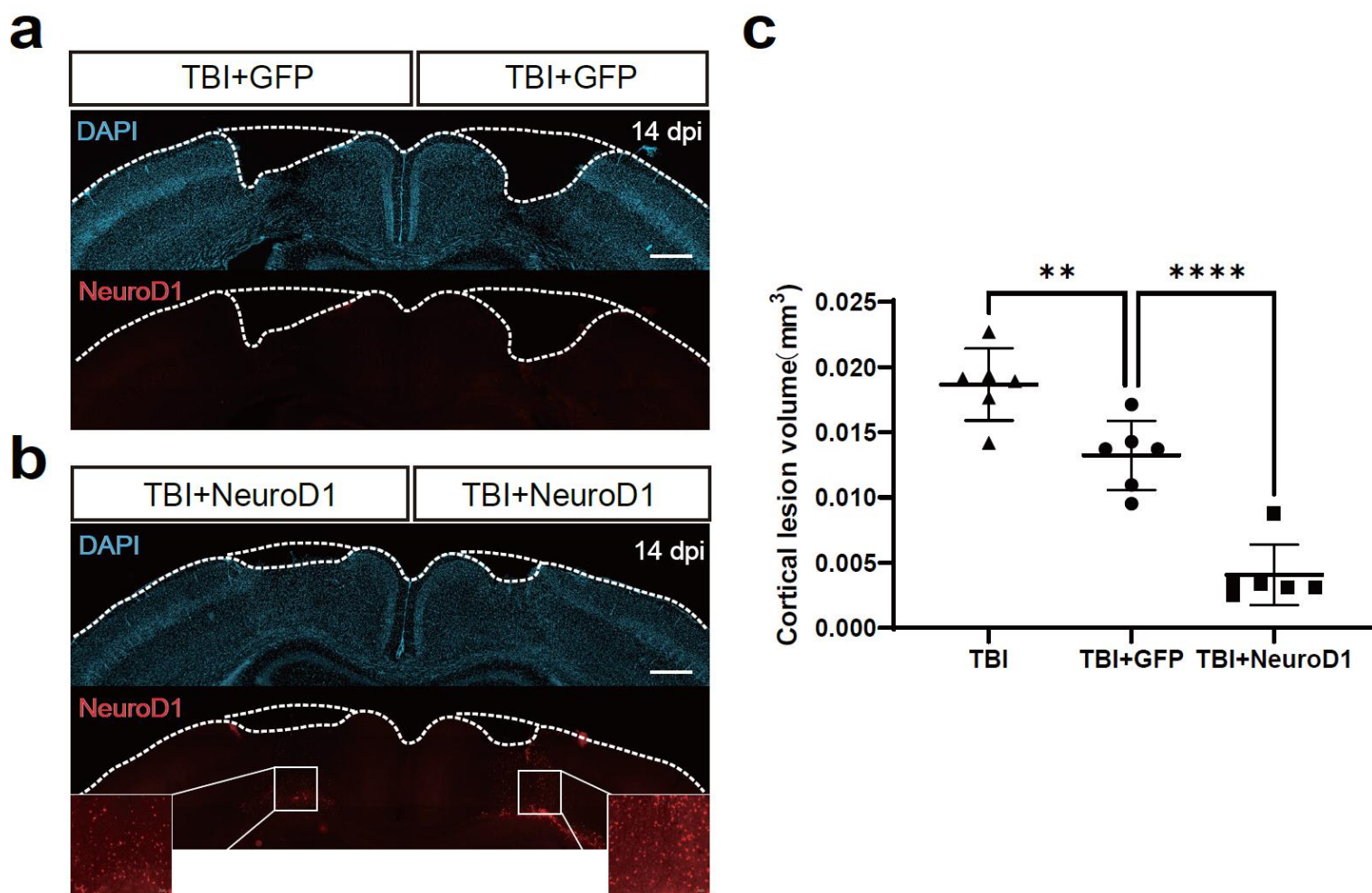
Supplementary Figures



Supplementary Figure 1: NeuroD1 promotes brain tissue repair and attenuates microglial activation at 7 days post-treatment following TBI

(a) Representative coronal sections obtained at 7 days post-treatment (dashed lines delineate injury boundaries). Inset shows NeuroD1 expression in the lesion area. Left: AAV9 GFAP::GFP; right: AAV9 GFAP::NeuroD1-GFP. DAPI (blue); NeuroD1 (red). Scale bar, 500 μ m, inset 50 μ m. **(b)** Quantification of cortical lesion volume from brain slices at 7 days post-treatment (n = 6; mean $\pm$ s.d.; one-way ANOVA with Tukey's post-hoc test; *P < 0.05, **P < 0.01). **(c)** Microglial activation in the lesion area at 7 days post-treatment. Iba-1 (purple). Scale bar, 500 μ m. **(d)** Quantification of Iba-1⁺ signal intensity at 7 days post-treatment, and TBI+NeuroD1 at 14 days post-treatment (n = 5-8; mean $\pm$ s.d.; one-way

ANOVA with Tukey's post-hoc test; * $P < 0.05$, **** $P < 0.0001$).



Supplementary Figure 2: Bilateral tissue damage and repair in TBI + GFP versus TBI + NeuroD1 groups at 14 days post-treatment following TBI

(a) Representative coronal sections obtained at 14 days post-treatment (dashed lines delineate injury boundaries). Left: AAV9 GFAP::GFP; right: AAV9 GFAP::GFP. DAPI (blue); NeuroD1 (red). Scale bar, 500 μ m. **(b)** Representative coronal sections obtained at 14 days post-treatment (dashed lines delineate injury boundaries). Insets show NeuroD1 expression in the lesion area. Left: AAV9 GFAP::NeuroD1-GFP; right: AAV9 GFAP::NeuroD1-GFP. DAPI (blue); NeuroD1 (red). Scale bar, 500 μ m, insets 50 μ m. **(c)** Quantification of cortical lesion volume from brain slices at 14 days post-treatment ($n = 6$; mean $\pm$ s.d.; one-way ANOVA with Tukey's post-hoc test; ** $P < 0.01$, **** $P < 0.0001$).

Supplementary Tables

Down (Normal) -Up (TBI+GFP) -Down (TBI+ NeuroD1) (FDR: Normal vs TBI+GFP, TBI+GFP vs TBI+NeuroD1)
mt-Rnr1 (0,1.08834073246331E-100)
GFP (9.27847623892011E-236,1.02759854640508E-07)
Cmss1 (0, 5.59360274233486E-122)
Lsamp (7.11322132973762E-172,9.13417779658289E-69)
Gpc5 (3.93149484858937E-107,2.09081007258284E-34)
Up (Normal) -Down (TBI+GFP) -Up (TBI+ NeuroD1) (FDR: Normal vs TBI+GFP, TBI+GFP vs TBI+NeuroD1)
Actb (0,5.69819522332762E-97)
Cnp (3.75569993618593E-168,3.6417492209067E-67)
Rps14 (5.24434549705114E-216,4.91454214920004E-50)
Cox6b1 (2.69611589287443E-221 ,1.68615688879645E-47)
Ndufb6 (2.71504100938691E-213,1.40675204428961E-44)

Supplementary Table 1: TBI-induced DEGs (TBI+GFP vs. Normal) that were reversed by NeuroD1 intervention (TBI+NeuroD1 vs. TBI+GFP), presented with FDR-adjusted effect sizes.

Down (Normal) -Up (TBI+GFP) -Down (TBI+ NeuroD1) (FDR: Normal vs TBI+GFP, TBI+GFP vs TBI+NeuroD1)
Cmss1 (4.78E-23,0.0000118)
mt-Rnr1 (4.82E-55,0.00000602)
mt-Rnr2 (2.08E-57,0.006802068)
Mbp (0.000327767,0.004747438)
Up (Normal) -Down (TBI+GFP) -Up (TBI+ NeuroD1) (FDR: Normal vs TBI+GFP, TBI+GFP vs TBI+NeuroD1)
Igfbp2 (4.05E-27,0.000000000903)
Chchd10 (7.01E-18,0.00000155)
Zfp871 (0.000000000448,0.00000154)
Ndufc2 (7.2E-20,0.0000154)
Ezr (3.19E-19,0.000178554)

Supplementary Table 2: DEGs in astrocyte subcluster 3 after TBI (TBI+GFP vs. Normal) and subsequently reversed by NeuroD1 intervention (TBI+NeuroD1 vs. TBI+GFP), with FDR-adjusted effect sizes.

Down (Normal) -Up (TBI+GFP) -Down (TBI+ NeuroD1) (FDR: Normal vs TBI+GFP, TBI+GFP vs TBI+NeuroD1)
GFP (0.0000838,0.035732467)
Cmss1 (9.71E-28,0.0000521)
mt-Rnr1 (4.55E-57,0.0000019)
mt-Rnr2 (3.44E-57,0.001030905)
Up (Normal) -Down (TBI+GFP) -Up (TBI+ NeuroD1) (FDR: Normal vs TBI+GFP, TBI+GFP vs TBI+NeuroD1)
Fabp7 (3.18E-23,0.00000403)
Olig1 (3.27E-22,0.001030905)
Slitrk2 (0.0000000000000000336,0.001030905)
Ostc (1.16E-22,0.0000000000000000116)
Igfbp2 (1.46E-21,0.0000121)

Supplementary Table 3: DEGs in astrocyte subcluster 6 after TBI (TBI+GFP vs. Normal) and subsequently reversed by NeuroD1 intervention (TBI+NeuroD1 vs. TBI+GFP), with FDR-adjusted effect sizes.