

Figure S1: Microglia at rostral or caudal 1 mm showed no difference in the mumber and morphology.

(A) Confocal images of microglia in spinal cord sections taken both rostral (R) and caudal (C) 1 mm to the lesion epicenter. Scale bars: 100 µm. (B) Quantification of microglia branch in spinal cord sections taken both rostral (R) and caudal (C) 1 mm to the lesion epicenter 4 weeks after astroglia transplantation (n = 5 per group). One-way ANOVA followed by Tukey's test. (C) Quantification of total number of Iba1⁺ microglia per mm^2 of tissue in spinal cord sections taken both rostral (R) and caudal (C) to the lesion epicenter at 4 weeks after astroglia transplantation (n = 5 per group). Oneway ANOVA followed by Tukey's multiple comparisons test. (D) Quantification of the total number of Iba1⁺ microglia per mm² of tissue in spinal cord sections taken both rostral (R) and caudal (C) to the lesion epicenter at 3-56 days after injury (n = 5 per group) (E) Quantification of the total number of Iba1⁺ microglia per mm² of tissue in spinal cord sections taken both rostral (R) and caudal (C) to the lesion epicenter at 3-56 days after NPC-Astro transplantation (n = 5 per group). (F) Quantification of the total number of Iba1⁺ microglia per mm² of tissue in spinal cord sections taken both rostral (R) and caudal (C) to the lesion epicenter at 3-56 days after Olig2PC-Astro transplantation (n = 5 per group). Two-way ANOVA followed by Tukey's multiple comparisons test. Data are mean \pm SD; n.s., not significant.



day(s) post injury

Figure S2: Grafted astroglia did not promote the increase of microglia.

(A-C) Representative images showing GFAP (red) and Iba1 (green) labeling in spinal cord sagittal sections at 4 weeks post-transplantation. A1, B1, C1 are higher magnification images of boxed areas in A-C. Scale bars: 200 μ m (A-C), 20 μ m (A1-C1). (D) Quantification of total Iba1⁺ microglia (n = 5 per group). (E) Quantification of Iba1⁺ microglia in the lesion area (n = 5 per group). (F) Western blotting for Iba1 expression in the lesion area at 3 to 56 days after transplantation. (G) Quantification of Iba1 expression at 3 to 56 days (n = 3 per group). D, E, G: Two-way ANOVA followed by Tukey's multiple comparisons test. Data are mean \pm SD; n.s., not significant.



Figure S3: Grafted astroglia did not promote microglia proliferation.

(A-D) Representative images showing ki67 (red) and Iba1 (green) staining in the lesion center at 4 weeks post-transplantation. Scale bars: 20 µm. (E) Quantification of the number of actively proliferating microglia (Iba1⁺ Ki67⁺ cells) at 28 days after astroglia transplantation (n = 5 per group). (F) Percentage of Iba1⁺ microglia undergoing proliferation 28 days after astroglia transplantation (n = 5 per group). (G, I, K, M) Quantification of the number of actively proliferating microglia (Iba1⁺ Ki67⁺ cells) at 3, 7, 14, 56 days after astroglia transplantation (n = 5 per group). (H, J, L, N) Percentage of Iba1⁺ microglia undergoing proliferation 3, 7, 14, 56 days after astroglia transplantation (n = 5 per group). Two-way ANOVA followed by Tukey's multiple comparisons test. Data are mean ± SD; n.s., not significant.



Figure S4: Grafted astroglia promote anti-inflammatory polarization of microglia. (A-C) Representative images showing Iba1 (green) and iNOS (red) labeling in spinal cord sagittal sections at 4 weeks post-transplantation. Scale bars: 200 µm (A-C), 20 µm (A1-C1). (D) Quantification of total iNOS⁺ microglia (n = 5 per group). (E) Quantification of iNOS⁺ microglia in the lesion area (n = 5 per group). (F-H) Representative images showing Iba1 (green) and Arg1 (red) labeling in spinal cord sagittal sections at 4 weeks post-transplantation. Scale bars: 200 µm (F-H), 20 µm (F1-H1). (I) Quantification of total iNOS⁺ microglia (n = 5 per group). (J) Quantification of iNOS⁺ microglia in the lesion area (n = 5 per group). (J) Quantification of iNOS⁺ microglia in the lesion area (n = 5 per group). (J) Quantification of iNOS⁺ microglia in the lesion area (n = 5 per group). (J) Quantification of iNOS⁺ microglia in the lesion area (n = 5 per group). (J) Quantification of iNOS⁺ microglia in the lesion area (n = 5 per group). (J) Quantification of iNOS⁺ microglia in the lesion area (n = 5 per group). (J) Quantification of iNOS⁺ microglia in the lesion area (n = 5 per group). D, E, I, J: Two-way ANOVA followed by Tukey's multiple comparisons test. Data are mean \pm SD; *p < 0.05, **p < 0.01, ***p < 0.001, n.s., not significant.