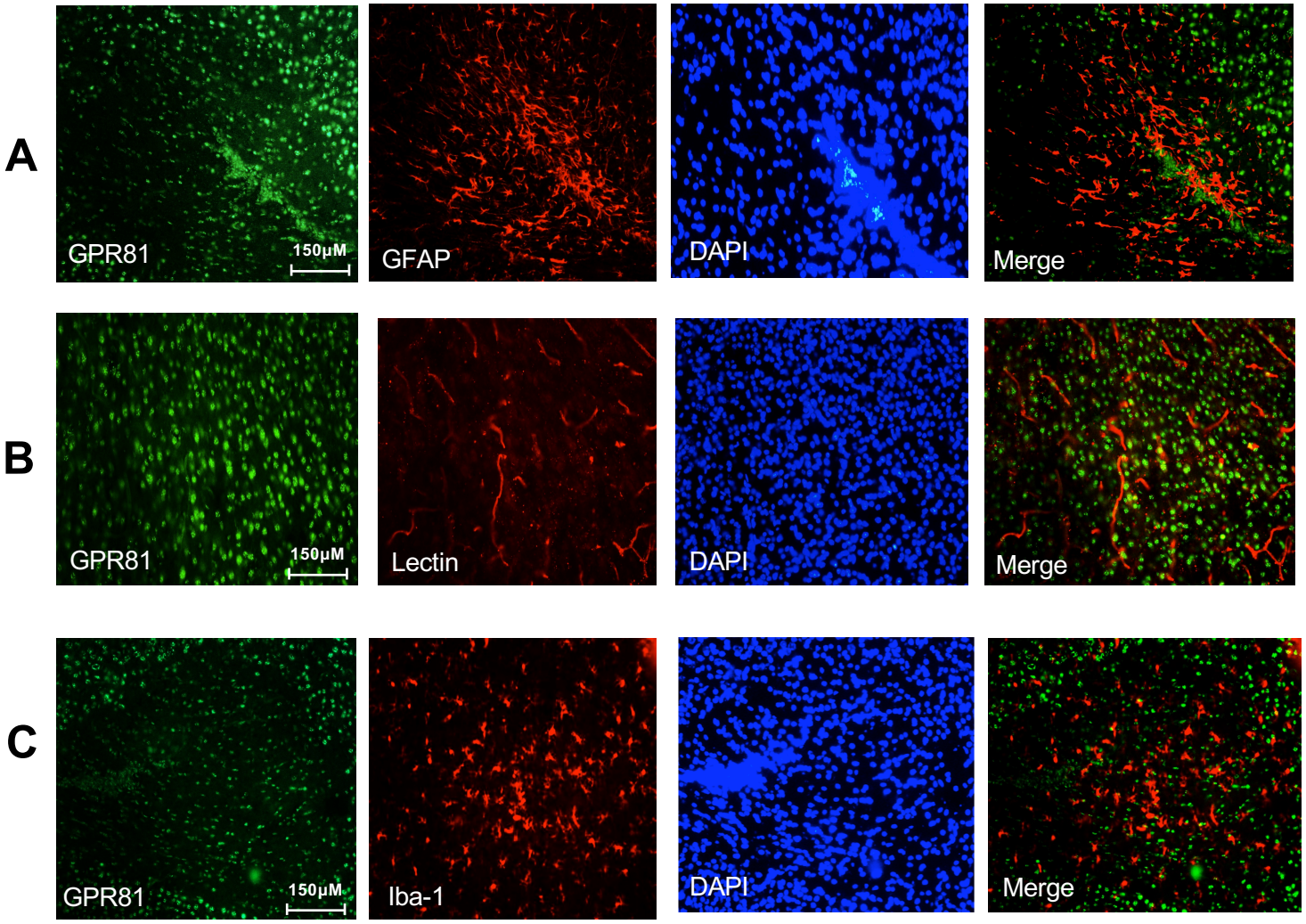
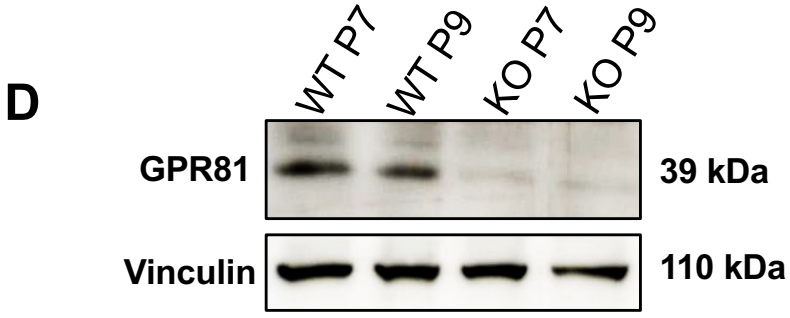
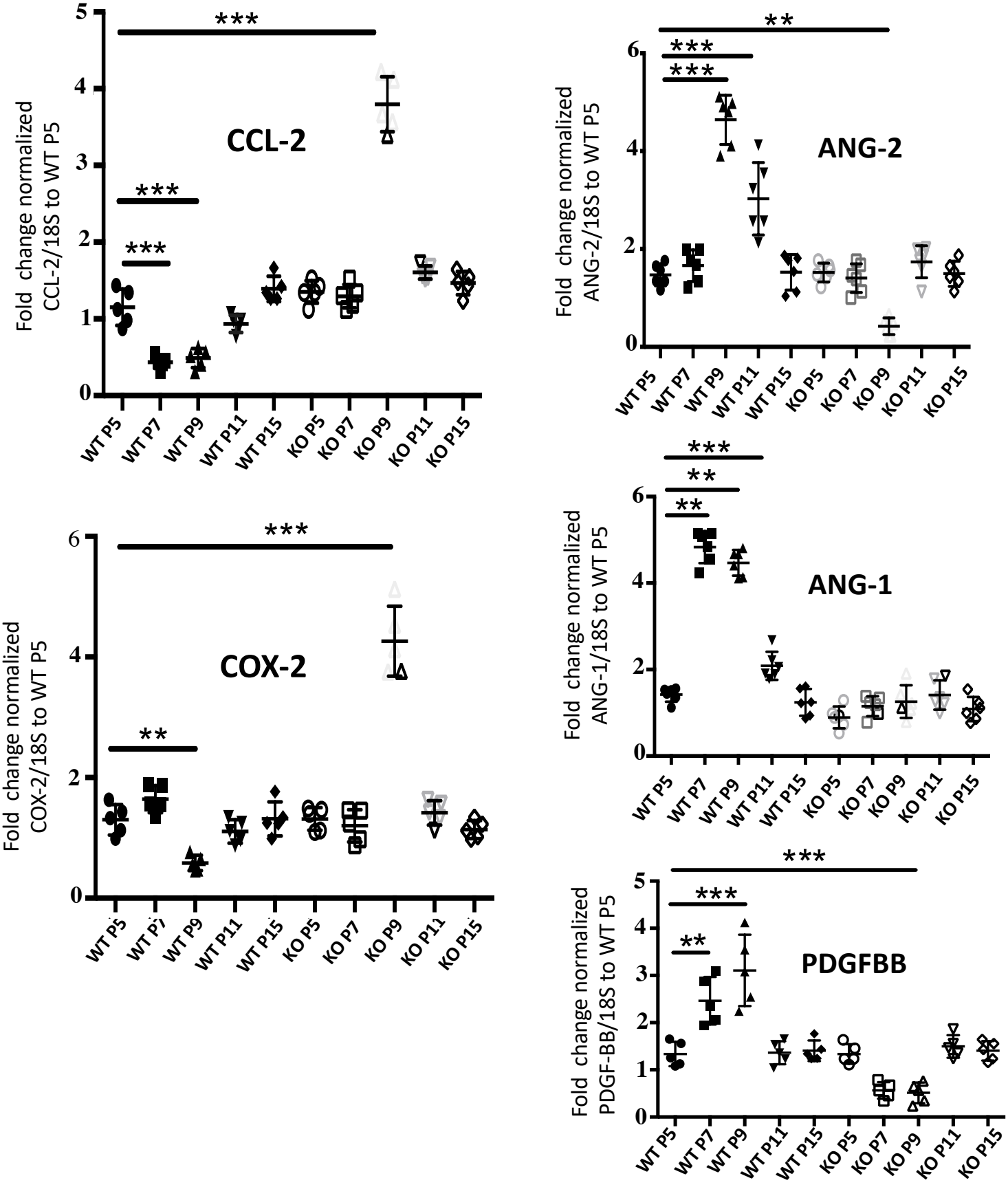
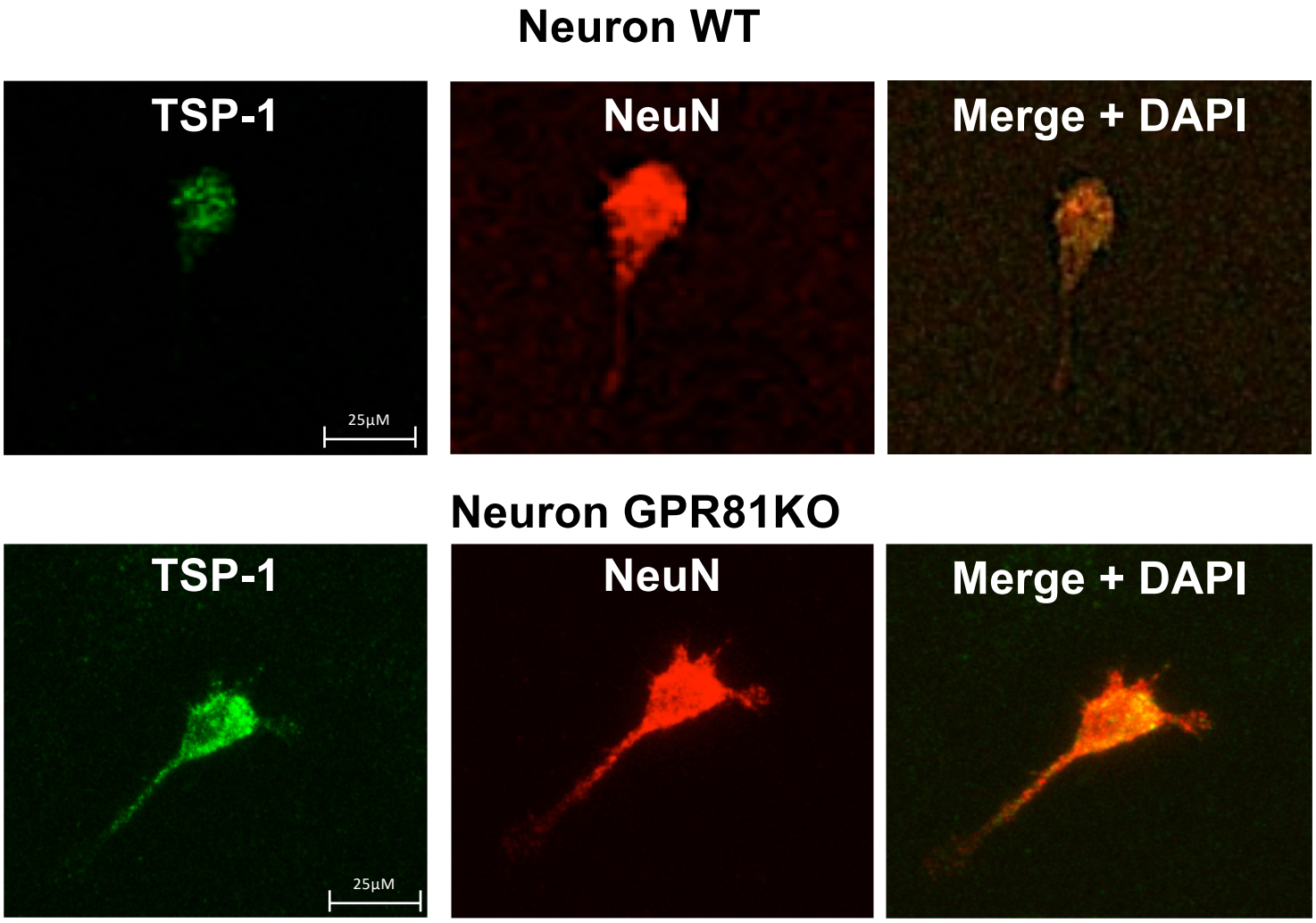




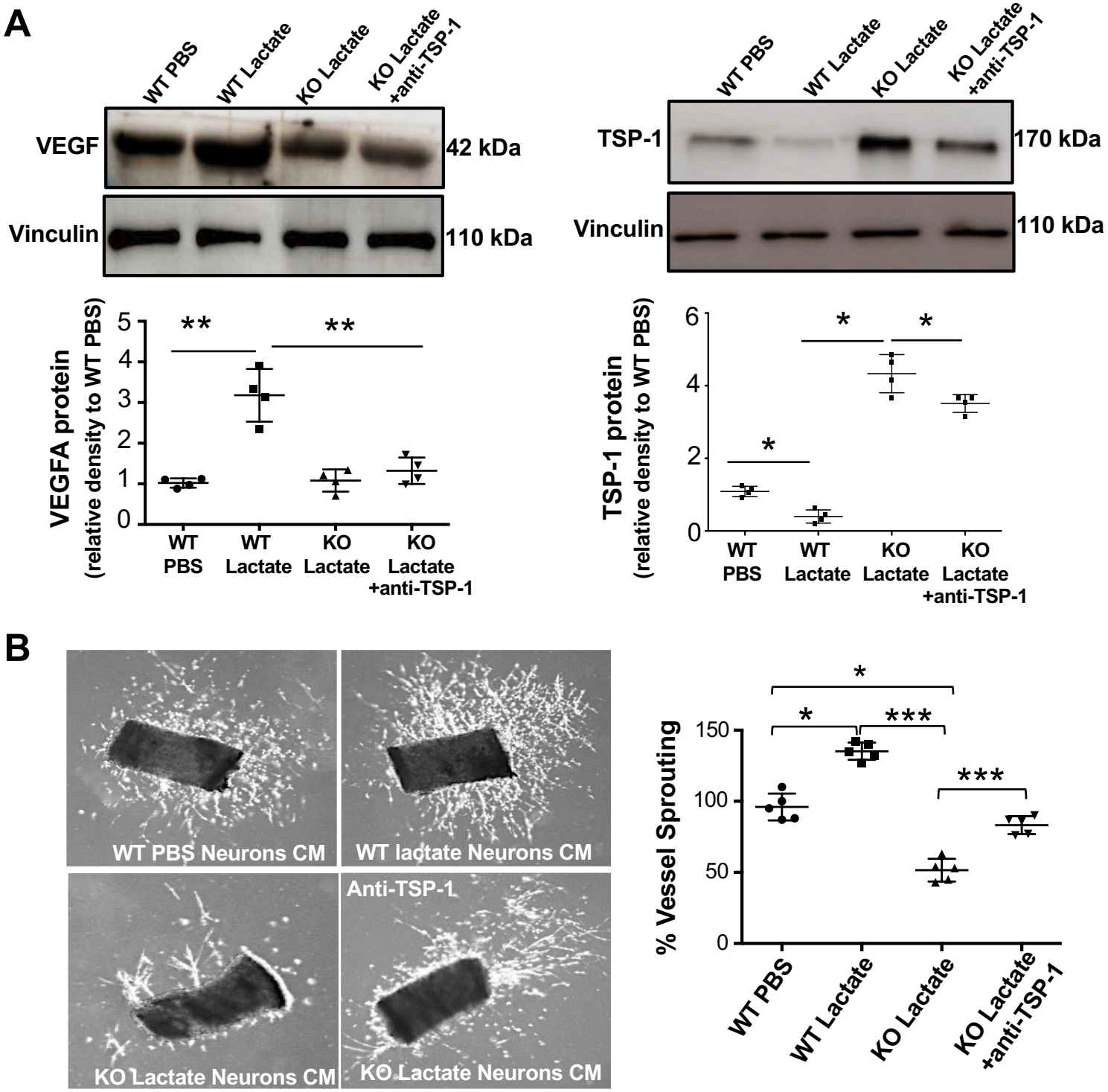
Supplementary Figure 1. Lactate receptor GPR81 is absent in astrocytes, microglia and endothelium in cerebral cortex. Representative confocal images of cerebral cortex sections from 3 animals showing a lack of colocalization of GPR81 (green) with **A**) GFAP (red), a specific marker for astrocytes, **B**) Lectin (red), an endothelium marker, and **C**) Iba-1 (red), an specific marker for microglial cells. Nuclei were counterstained with 4',6-diamidino-2-phenylindole (DAPI) and merged images (yellow) show colocalization. Scale bar, 150 μm. **D**) Western blot analysis showing immunoreactivity to GPR81 (39 kDa) in brain homogenates from WT, but absent in brains of GPR81-null-mice. Vinculin (110 kDa) was used as an internal control.



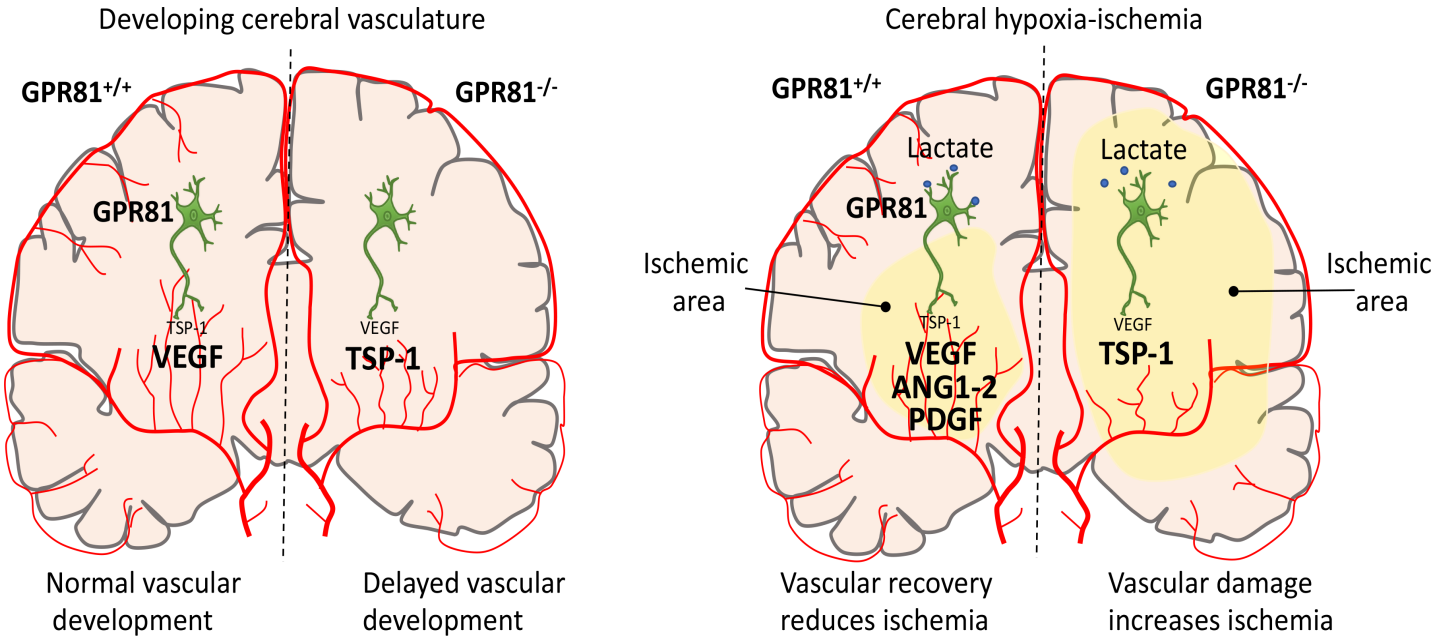
Supplementary Figure 2. Ontogenic expression of major angiogenic and proinflammatory factors in wild-type and GPR81-null mouse brain cortex. Real-time quantitative PCR analysis of Ang-1, Ang-2, PDGFBB, COX-2 and CCL-2 in WT mice and GPR81-null mice at different postnatal ages. Values represent mean ± SD; n = 5 samples per group. **p<0.01, ***p<0.001 compared with their respective control, one-way ANOVA followed by Dunnett's test for multiple comparison with control.



Supplementary Figure 3. TSP-1 is overexpressed in neurons from GPR81-null mice. Representative confocal images of isolated primary neurons from WT and GPR81-null mice showing colocalization of TSP-1 (green) with the specific neuronal marker NeuN (red). Nuclei were counterstained with 4',6-diamidino-2-phenylindole (DAPI) and the merged images (yellow) show the colocalization. Scale bar, 25μm.



Supplementary Figure 4. Lactate-triggered induction of VEGF-A and suppression of TSP-1 in hypoxic neurons elicits neovascularization *ex vivo*. (**A**) Western blot analysis of VEGF (42 kDa) and TSP-1 (170 kDa) protein levels in the conditioned media from WT- and GPR81-null mice-derived neurons stimulated with lactate (10 mmol/L) or vehicle (PBS). Vinculin (110 kDa) evaluated in the same neuronal homogenates were the condition media was collected, was used as an internal control. Densitometry quantification is illustrated in the histograms. Data are expressed as mean ± SD; n = 3 experiments per group; *p<0.05, **p<0.01, compared to corresponding WT PBS, one-way ANOVA followed by Dunnett's test for multiple comparison with control. (**B**) Representative microvascular sproutings of Matrigel-embedded aortic explants treated for 96 hours with the conditioned media from hypoxic WT- and GPR81-null mice-derived primary neurons previously stimulated with lactate (10 mmol/L) or vehicle (PBS). Histograms represent the quantification of the aortic vessels sprouting vessel. Data are presented as mean ± SD; n = 5 aortic explants per group; *p<0.05, ***p<0.001 compared to WT PBS neurons, one-way ANOVA followed by Dunnett's test for multiple comparison with control.



Supplementary Figure 5. Brain schemes showing the role of neuronal GPR81 in normal vascular development and in cerebral hypoxia-ischemia. During development, GPR81 activation in the neurons triggers VEGF production that controls brain vascular development. In absence of GPR81, a significant delay in the development of brain vasculature is associated with augmented production of the anti-angiogenic factor TSP-1 (left image). During a cerebral hypoxia ischemia episode, lactate administration acting through GPR81 triggers the production of proangiogenic factors (VEGF, Ang-1 and 2, PDGF) facilitating healthy revascularization and diminishing ischemic area in wild-type mice. Conversely, in the absence of GPR81, lactate does not trigger an appropriated brain vascular restoration, thus aggravating the area of ischemic hypoxia, as elevated levels of neuronal TSP-1 are maintained (right image).