Supplemental information

Amlexanox inhibits cerebral ischemia-induced delayed astrocytic high-mobility group box 1 release and subsequent brain damage

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**Supplementary Fig. 1.** Amlexanox inhibits ischemia-induced HMGB1 release from astrocytes in the striatum and hippocampus of mouse brain. (A-D) Mice were subjected to 1 h tMCAO. (A) Immunostaining of GFAP (astrocyte marker) and HMGB1 (GFAP, green; HMGB1, red) using brain coronal sections was performed at 3 h after tMCAO. Following amlexanox (Amx) (10 µg/5 µL) or PBS (ischemic vehicle) treatment (i.c.v.) at 24 h after tMCAO, immunostaining of GFAP and HMGB1 was performed at 27 h after tMCAO. (B) Quantitative analysis of HMGB1-positive astrocytes in striatum in the brain at 3 and 27 h after tMCAO. Data are means ± SEM. One-way ANOVA, Dunnett’s multiple comparison tests, *p < 0.05 versus untreated mice, #p < 0.05 versus ischemic vehicle at 27 h. (C) Immunostaining of GFAP and HMGB1 in the stratum radiatum (Str
rad) of hippocampus at 3 and 27 h after tMCAO in ischemic vehicle and Amx post-treated (24 h) mice. (D) Quantitative analysis of HMGB1-positive astrocytes in the Str rad at 3 and 27 h after tMCAO. Data are means ± SEM. One-way ANOVA, Dunnett’s multiple comparison tests, *p < 0.05 versus untreated mice, #p < 0.05 versus ischemic vehicle at 27 h. n = 3 in each group.