

**Title: CCL5 via GPX1 activation protects hippocampal memory function after mild traumatic brain injury**

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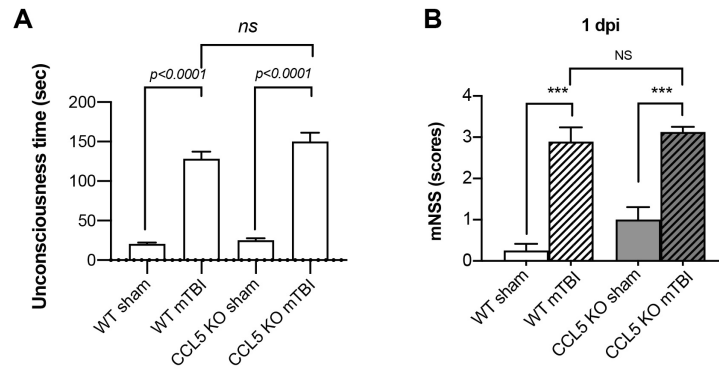
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**Supplementary Information:**

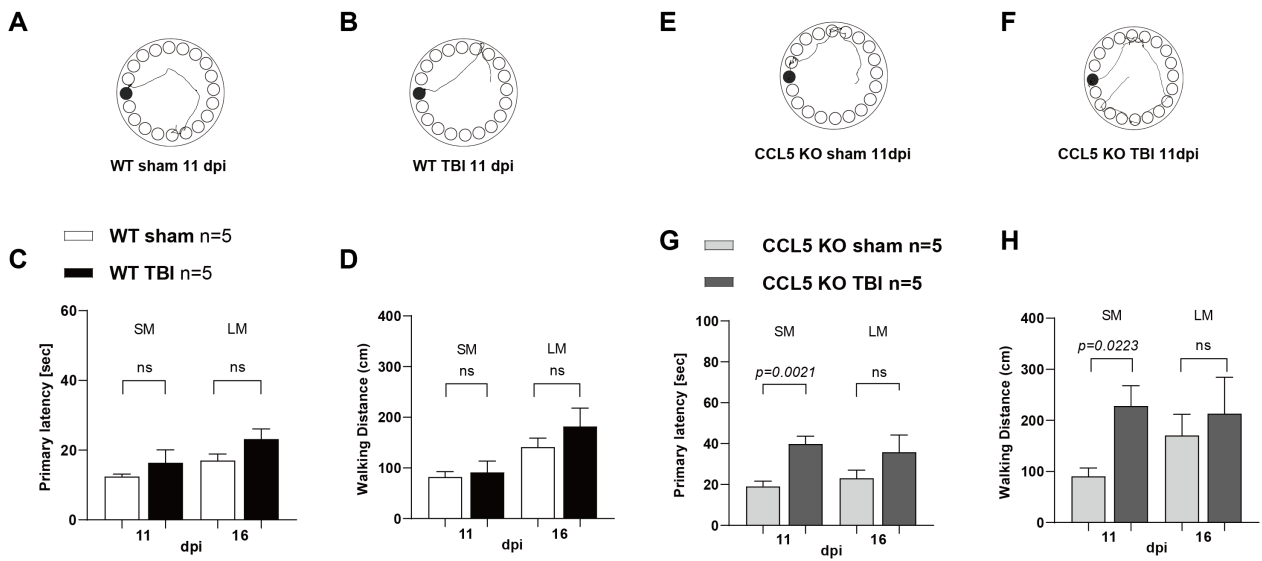
**Includes 6 Supplementary Figure legends, 6 Supplementary Figures**

**Supl. Figures:**

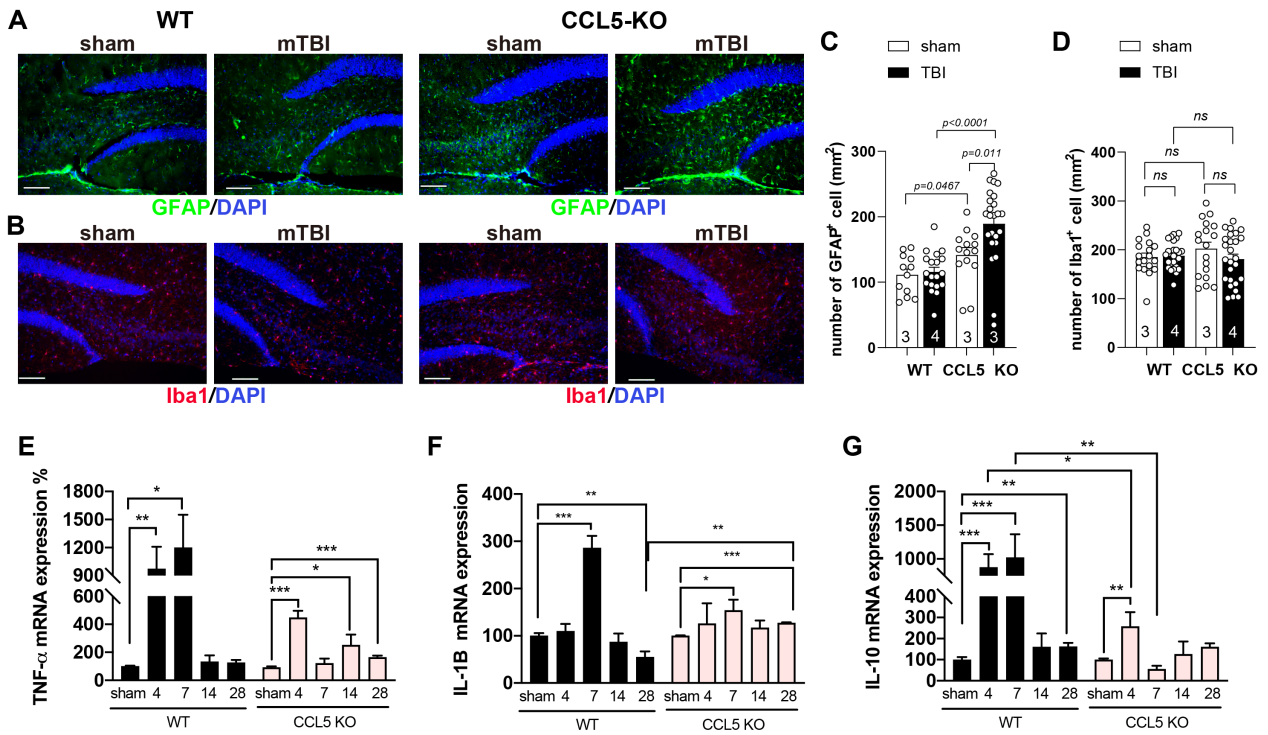


**Supl. Fig. 1: The unconsciousness times and mNSS score of WT and CCL5-KO mice after mTBI.**

(A) The unconsciousness time in WT and CCL5-KO sham group and weight-drop induced mTBI group. (B) The modified neurological severity score (mNSS) was performed at 1 day post injury. (\*,  $p < 0.05$ ; \*\*,  $p < 0.01$ ; \*\*\*,  $p < 0.001$ . ns: no significant difference. Data was analyzed by *t*-test and presented as mean  $\pm$  S.E.M).

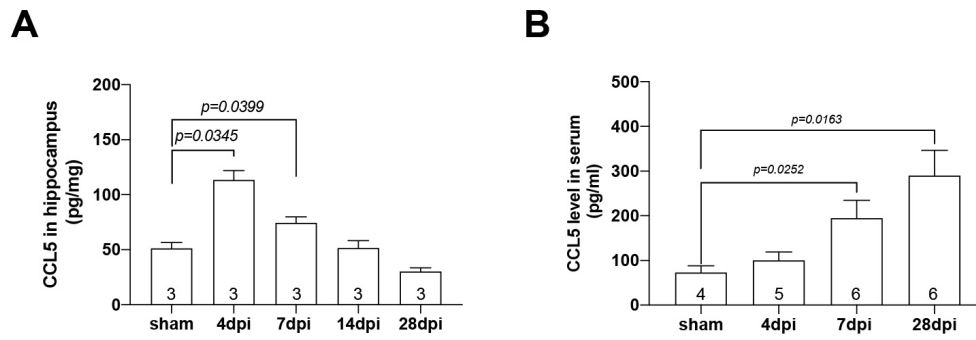


**Supl. Fig. 2: Spatial memory changes after mTBI in WT and CCL5 KO mice in the Barnes maze.** (A, B, E, F) Representative walking tracks in the Barnes maze in WT and CCL5-KO mouse sham and mTBI groups 11 days after injury. The primary latency and walking distance of WT (C, D) and CCL5-KO mice (G, H) sham and mTBI groups at 11 and 16 dpi. (Data was analyzed by *t*-test and presented as mean± S.E.M).

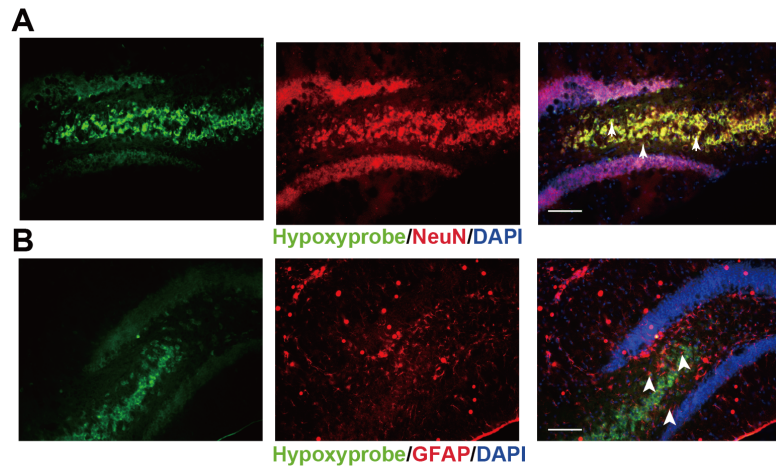


**Supl. Fig. 3: Immune cell and chemokine activations in mouse hippocampus after brain injury.**

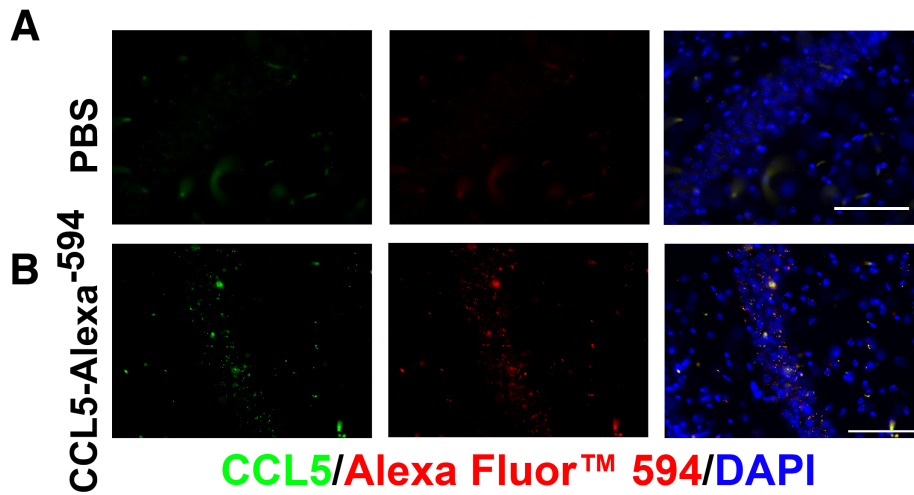
(A) Astrocyte density was labeled by GFAP (green) in sham and mTBI WT and CCL5-KO mice. (C) Quantification of GFAP labeled cells from 3~4 mice in each group. (B) Immunostaining of microglia with specific marker – Iba1 (red). DAPI labeled nucleus in blue in A, B. (D) Quantification of Iba1 positive cells from 2~3 mice in each group. Scale bar=100 $\mu$ m. mRNA expression of inflammatory chemokines TNF- $\alpha$  (E), IL-1 $\beta$  (F) and IL-10 in WT and CCL5-KO mouse hippocampus after brain injury. (\*,  $p < 0.05$ ; \*\*,  $p < 0.01$ ; \*\*\*,  $p < 0.001$ . Data was analyzed by  $t$ -test and presented as mean  $\pm$  S.E.M)



**Supl. Fig. 4: The levels of CCL5 in WT mouse hippocampus and serum after injury.** The concentration of CCL5 in sham mouse hippocampus (A) and serum (B) and mice with TBI after 4, 7, 14 and 28 days was analyzed by ELISA. (Data was analyzed by *t*-test between sham and after injury groups.)



**Supl. Fig. 5: Hypoxyprobe double labeling with NeuN and GFAP in mouse hippocampus.** (A) Hypoxic cells in mouse hippocampus were labeled by hypoxyprobe (green) which was co-labeled with NeuN (Red) in (A) or GFAP (Red) in (B). DAPI (blue) labeled the nucleus. Scale bar=100 $\mu$ m.



**Supl. Fig. 6: The distribution of Alexa-594 conjugated CCL5 in mouse hippocampus through intranasal delivery. (B) Recombinant CCL5 was labeled by Alexa-594 and delivered into CCL5-KO mouse brain. The localization of CCL5 in mouse hippocampus through intranasal delivery was confirmed by CCL5 specific antibody labeling (green) and conjugated protein - Alexa-594 (red fluorescence). Nucleus were labeled by DAPI (Blue). (A) Mouse receiving PBS was taken as control. Scale bar=100µm.**