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## Supplemental information

# Paradoxical effects of posterior intralaminar thalamic calretinin neurons on hippocampal seizure via distinct downstream circuits

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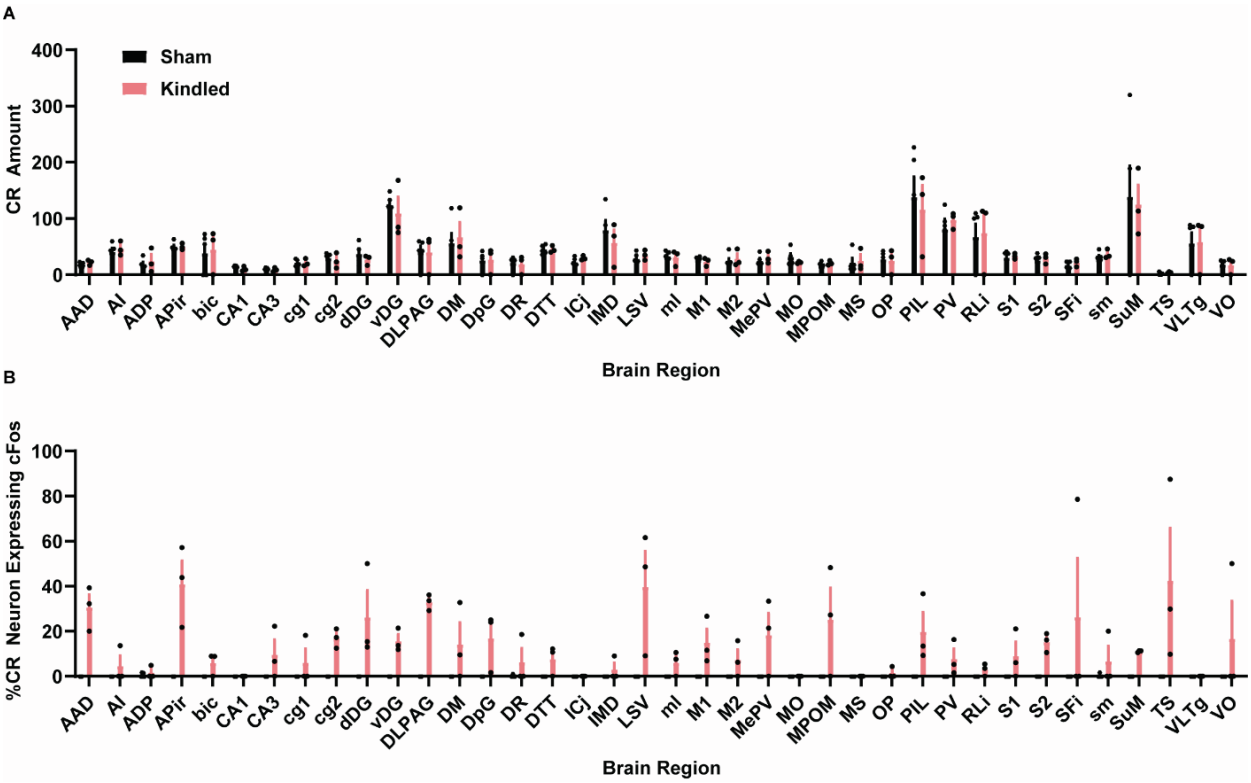


**Figure S1 Activation of CR neurons in different brain regions after hippocampal seizure.** (A and B) Average number of CR neurons (A) and cFos$^{+}$ CR neurons (B) in different brain regions of WT sham and kindling-induced epilepsy model (n = 5 for Sham group, n = 3 for Kindled group). Data are represented as mean ± SEM. AAD, anterior amygdaloid area (dorsal). AI, agranular insular cortex. ADP, anterodorsal preoptic nucleus. APir, amygdalopiriform transition area; bic, brachium of the inferior colliculus; CA1, field of CA1 of hippocampus; CA3, field of CA3 hippocampus; cg1, cingulate cortex area 1; cg2, cingulate cortex area 2; dDG, dorsal dentate gyrus ; vDG, ventral dentate gyrus; DLPAG, dorsolateral periaqueductal gray; DM, dorsomedial hypothalamic nucleus; DpG, deep gray layer of the superior colliculus; DR, dorsal raphe nucleus; DTT, dorsal tenia tecta; ICj, islands of Calleja; IMD, intermediodorsal thalmic nucleus; LSV, lateral septal nucleus (ventral); ml, medial lemniscus; M1, primary motor cortex; M2, secondary motor cortex; MePV, medial amygdaloid nucleus (posteroventral); MO, medial orbital cortex. MPOM, medial preoptic nucleus; MS, medial septal nucleus; OP, optic nerve layer of the superior colliculus; PIL, posterior intralaminar thalamic nucleus; PV, paraventricular thalamic nucleus; RLi, rostral linear nucleus of the raphe; S1, primary somatosensory cortex; S2, secondary somatosensory cortex; SFi, septofimbrial nucleus; sm, stria medullaris of the thalamus; SuM, supramammillary nucleus; TS, triangular septal nucleus; VLTg, ventrolateral tegmental area; VO, ventral orbital cortex. **Related to Figure 1.**

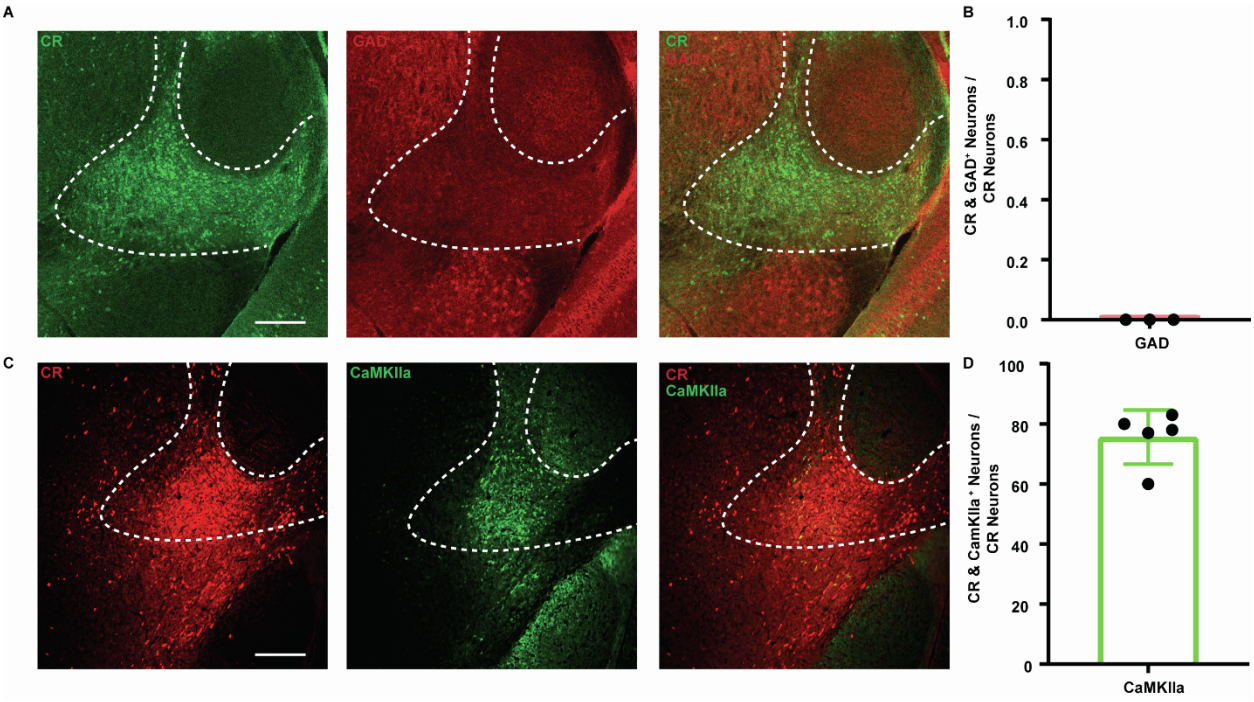


**Figure S2 CR neurons in the PIL are glutamatergic neurons.**
(A) Representative immunostaining images of CR neurons in the PIL region (green), GAD65/67 (red), and overlap of CR and GAD65/67 (Scale Bar: 200 μm).
(B) Statistical data for colocalization of CR neurons and GAD overlap (n=3).
(C) Representative immunostaining images of CR neurons in the PIL region (red), CaMKIIα (green), and overlap of PIL and CaMKIIα (Scale Bar: 200 μm).
(D) Statistical data for the percentage CR neurons that expressed CaMKIIα (n=5).
Data are represented as mean ± SEM. **Related to Figure 4.**

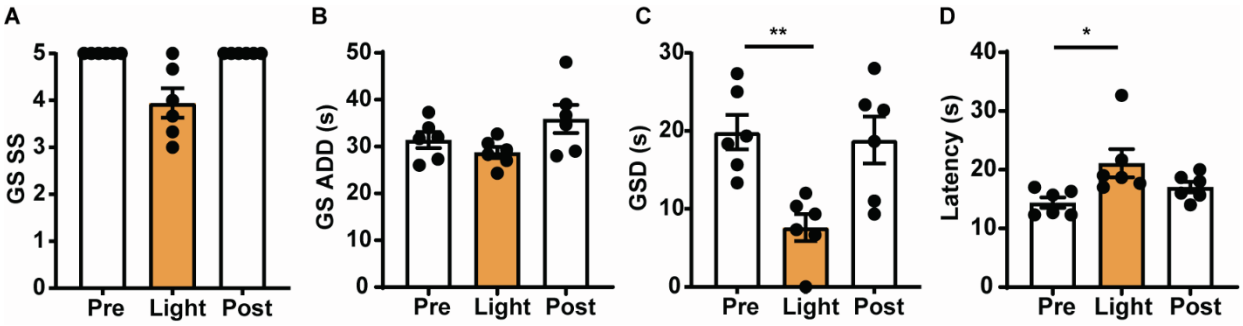


**Figure S3 Inhibition of PIL-LA CR circuit alleviates the severity of GS in fully kindled state.** (A-D) Effects of optogenetic inhibition of PIL-LA CR projections on seizure stage (SS, A), after-discharge duration (ADD, B), GS duration (GSD, C), and latency to GS (D) during hippocampal kindled seizure. (n = 6) *P<0.05, **P<0.01, one-way ANOVA. Data are represented as mean ± SEM. Related to Figure 5.

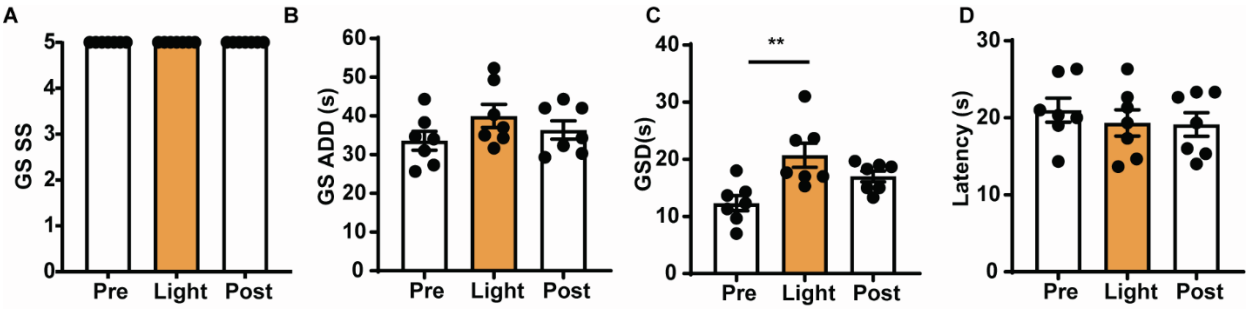


**Figure S4 Inhibition of PIL-ZI CR circuit aggravates the severity of GS in fully kindled state.** (A-D) Effects of optogenetic inhibition of PIL-ZI CR projections on seizure stage (SS, A), after-discharge duration (ADD, B), GS duration (GSD, C), and latency to GS (D) during hippocampal kindled seizure. (n = 7) **P<0.01, one-way ANOVA. Data are represented as mean ± SEM. **Related to Figure 6.**

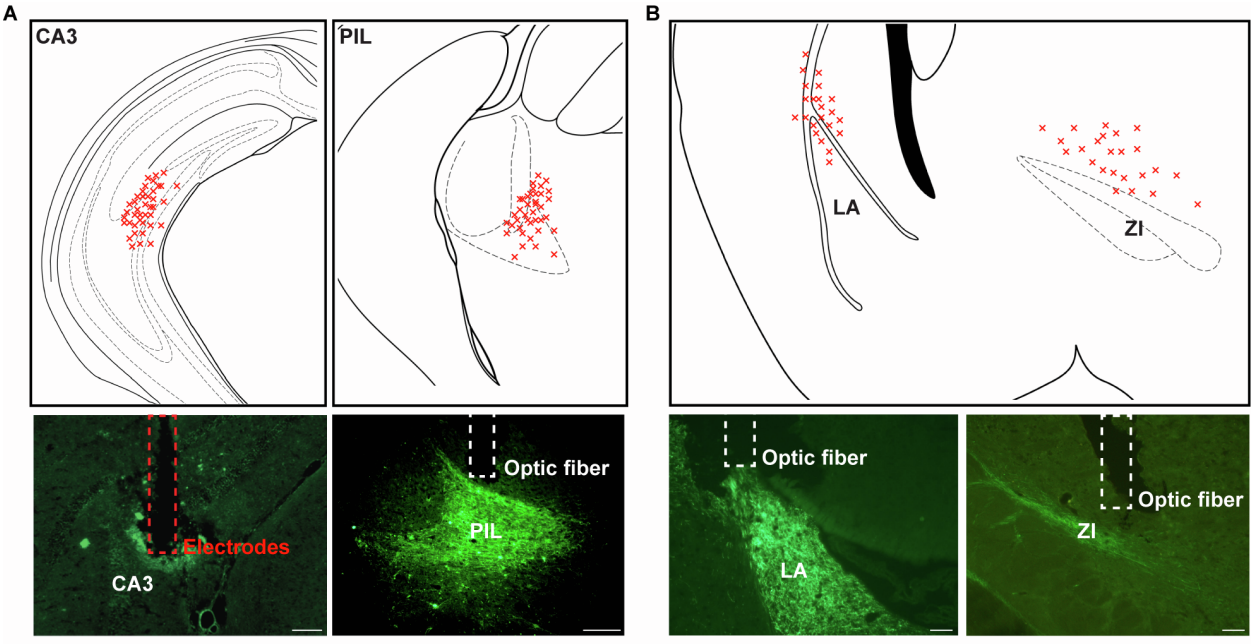


**Figure S5 Mapping the location of electrodes and optic fibers in optogenetic experiment.**
(A) Above, mapping the location of kindling electrodes in the CA3 and optic fibers in the PIL in optogenetic experiment in Figures 2 and 3. Below, representative images for the location of kindling electrodes in the CA3 and optic fibers in the PIL. **Related to Figure 2 and 3.**
(B) Above, mapping the optic fibers in the LA and ZI in optogenetic experiment in Figures 5 and 6. Below, representative images for the location of optic fibers in the LA and ZI. **Related to Figure 5 and 6.**

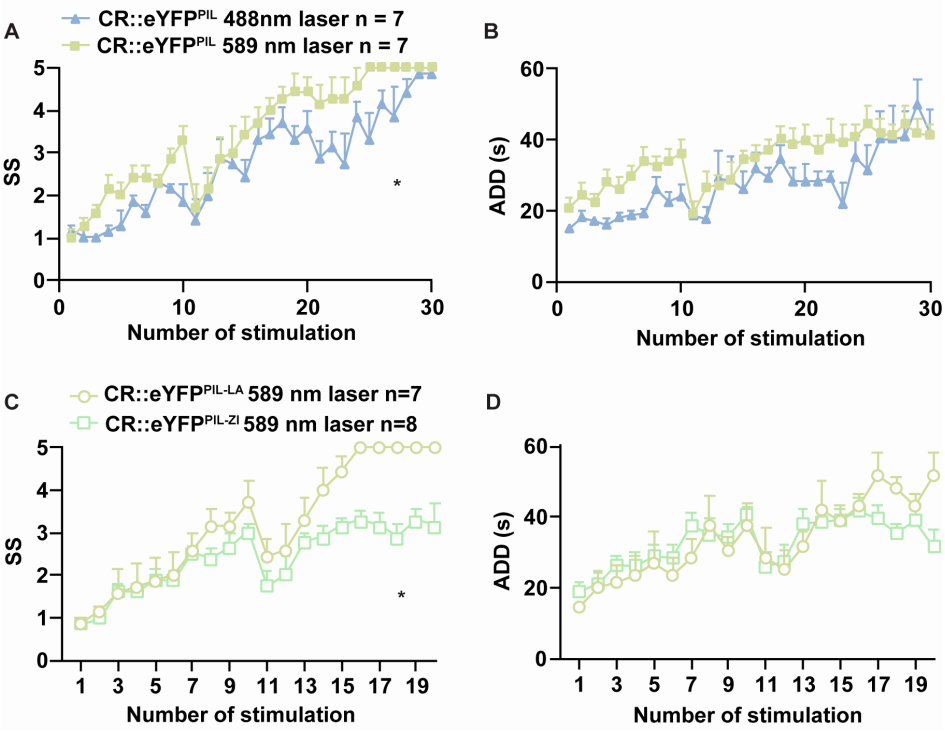


**Figure S6 Different epileptogenic susceptibility of control groups in optogenetic experiment.** (A and B) The development of seizure stage (SS, A) and after-discharge duration (ADD, B) in control groups of optogenetics experiment in Figures 2 and 3. *P<0.05, two-way ANOVA with repeated measures. Data are represented as mean ± SEM. **Related to Figure 2 and 3.**
(C and D) The development of SS (C) and ADD (D) in control groups of optogenetics experiment in Figures 5 and 6. *P<0.05, two-way ANOVA with repeated measures. Data are represented as mean ± SEM. **Related to Figure 5 and 6.**