

Fig. 1b

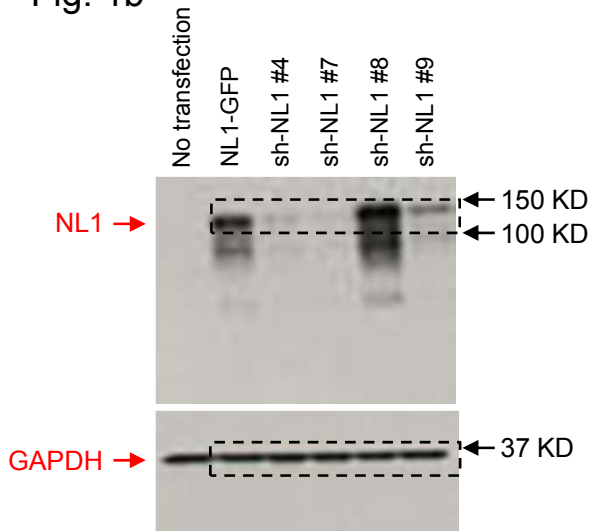


Fig. 1c

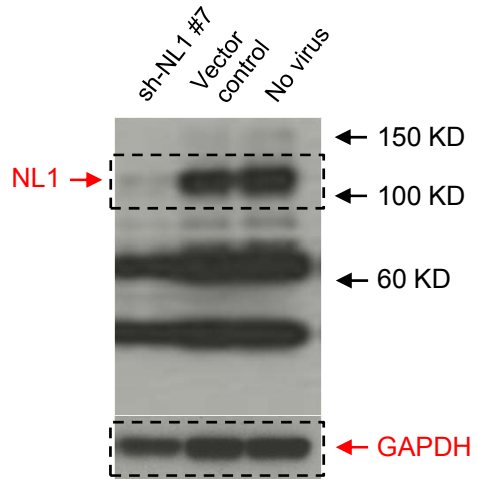
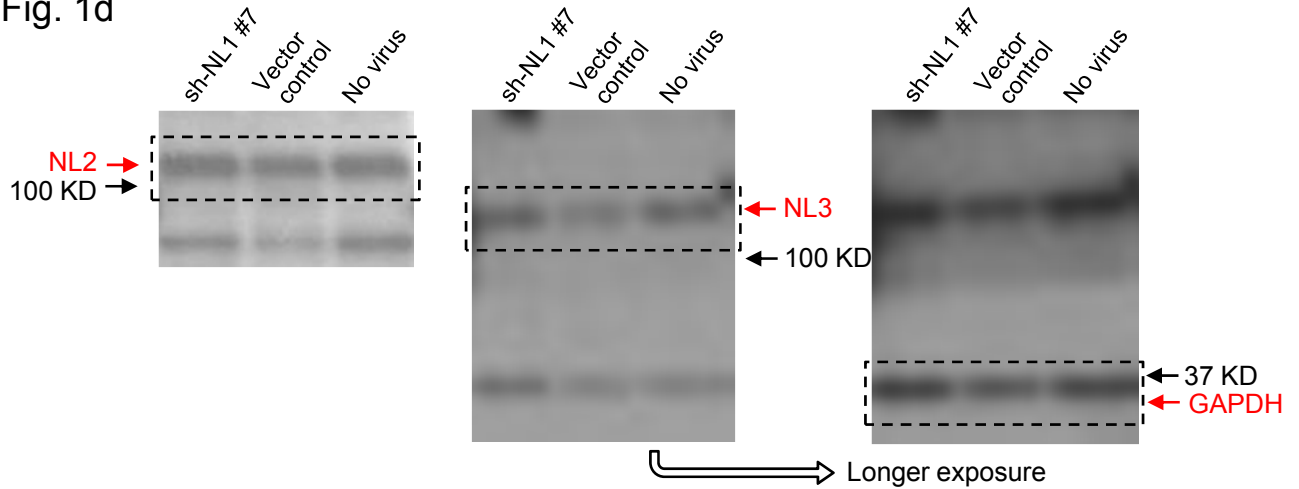
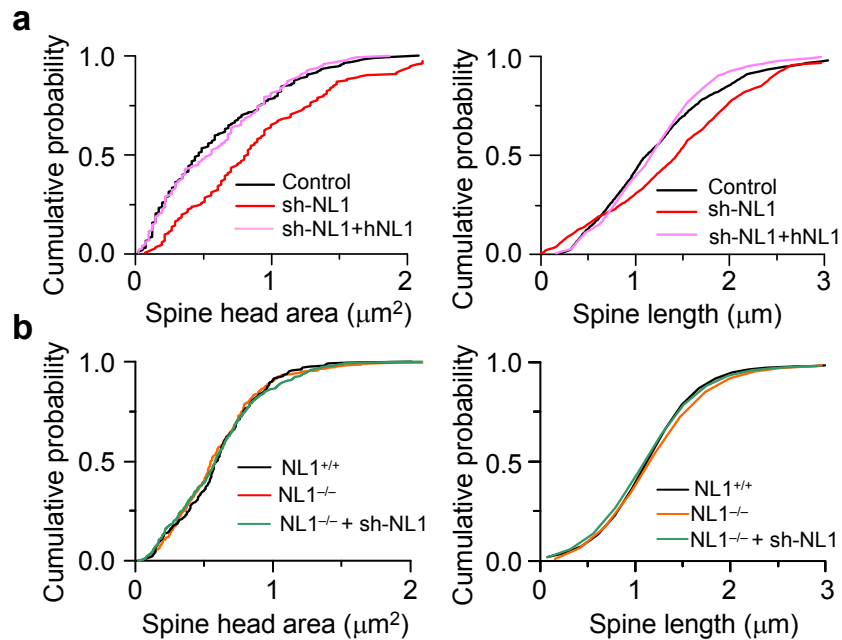


Fig. 1d



Supplementary Figure 1 Full images of western blots shown in the main figures

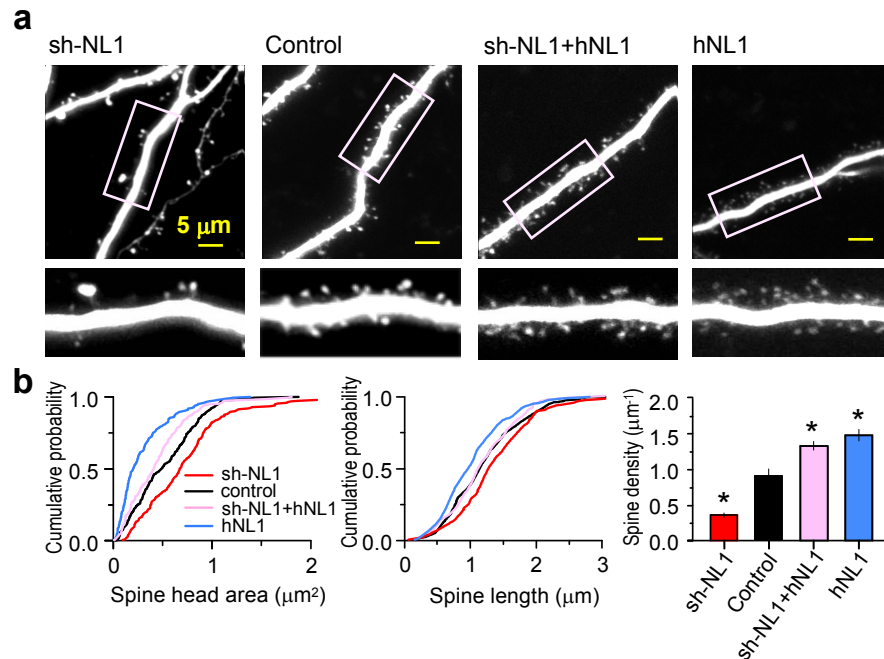
Bands within dashed line boxes are shown in Figure 1.



Supplementary Figure 2 Cumulative plots of spine head area and length

(a) Cumulative plots of spine head area and length from neurons transfected with the sh-NL1 #7 (sh-NL1), GFP (control), sh-NL1#7 and human NL1 (hNL1).

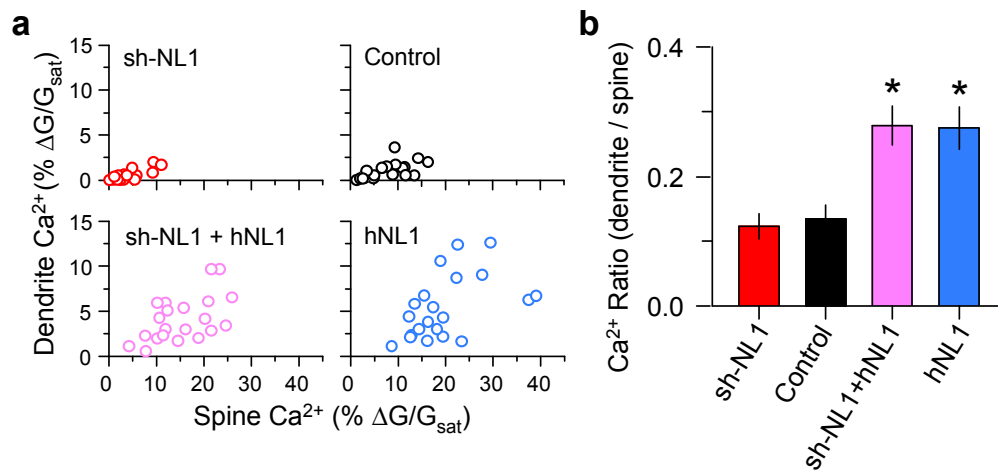
(b) Cumulative plots of spine head area and length from neurons of indicated genotypes.



Supplementary Figure 3 Postsynaptic expression of NL1 regulates spine morphology and density in CA1 pyramidal neurons in organotypic hippocampal slices

(a) Representative images of dendritic spines of hippocampal CA1 pyramidal neurons transfected with the sh-NL1, GFP (control), sh-NL1#7 and hNL1, or hNL1 alone. The areas indicated by the white boxes are shown at higher magnification below.

(b) Cumulative distributions of apparent spine head areas (*left*), lengths (*middle*), and average spine density (*right*) measured from hippocampal CA1 neurons in organotypic slice cultures of the genotypes shown in panel (a). Error bars represent s.e.m.

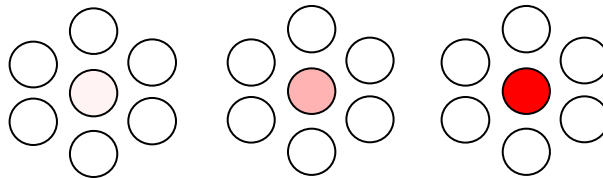


Supplementary Figure 4 Relative Ca²⁺ accumulation in the dendrite is increased by hNL1 overexpression

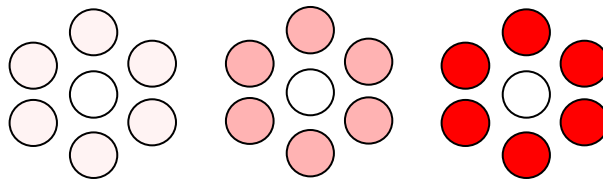
(a) Glutamate was photoreleased near the spine head and the Ca²⁺ transient peaks from the spine head and neighboring dendrite shaft were measured as described in Figure 2. Measurements from individual spines are plotted as open circles. Note that dendritic Ca²⁺ increased more steeply than spine Ca²⁺ when hNL1 was transfected (rescued and hNL1 groups).

(b) Average ratios of Ca²⁺ transient amplitudes in the dendrite relative to the spine, showing increased relative dendritic Ca²⁺ in rescued and hNL1 transfected neurons. Error bars represent s.e.m.

Absolute change
(cell-autonomous)



Relative change
(trans-cellular)



Supplementary Figure 5 Schematics of absolute and relative regulation of synapse number by NL1 expression

In the top, cell-autonomous model, changes in the level of expression of NL1 (red levels) in the center cell influence its own synapse number independent of the NL1 levels in the surrounding neurons. In the bottom, trans-cellular model, synapse number in the center cell is affected by the changes in NL1 levels of neighboring cells.