

Supplementary Figure 1. Immunofluorescence, as assessed by anti-PAR1b monoclonal antibody.
Representative images of mouse hippocampal neurons (DIV14) immunostained with anti-PAR1b monoclonal antibody (red) and anti-GFP antibody (green). Neurons were transfected with nonsilencing control vector (NC) or PAR1b knockdown vector (PAR1b KD) at DIV7. Each knockdown vector simultaneously expressed GFP. Scale bar, $10 \mu \mathrm{~m}$.


Supplementary Figure 2. Verification of PAR1b knockdown phenotype by an additional PAR1b knockdown vector.

Representative images of hippocampal neurons (DIV21) transfected with nonsilencing control vector (top panels), PAR1b knockdown vectors, Q3 (PAR1b KD Q3, middle panels) or \#1 (bottom panels) at DIV17. Scale bar, $10 \mu \mathrm{~m}$.

A


B




Supplementary Figure 3. GFP MAP2b overexpression in mature hippocampal neurons results in decreases size and density of dendritic spines.
Representative images of mouse hippocampal neurons (DIV21) transfected with GFP expressing vector (upper panels) or GFP-MAP2b expressing vector (lower panels) with tRFP expressing vector at DIV17. Neurons were immunostained with anti-tRFP antibody (red) and anti-GFP antibody (green). Scale bar, $10 \mu \mathrm{~m}$. (B) Quantification of mean dendritic protrusion width (left panel), length (middle panel) and density (right panel) in GFP overexpressing (GFP OE) cells and GFP-MAP2b overexpressing (GFP-MAP2b OE) cells. More than 600 dendritic protrusions from randomly selected cells ( $>12$ neurons) were analyzed for each condition. Bars show mean $\pm$ SEM. $* P<0.05$ by Student $t$ test.

