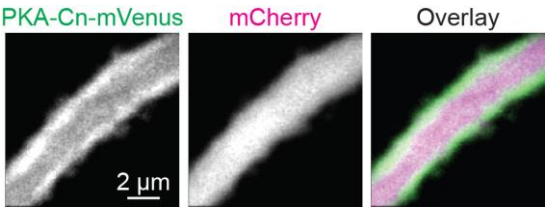

Fig. S1. Representative single color images and their overlay of PKA-Cn in neuronal dendrites.

Compared to the cytosolic marker mCherry, PKA-Cn was more enriched on the membrane.

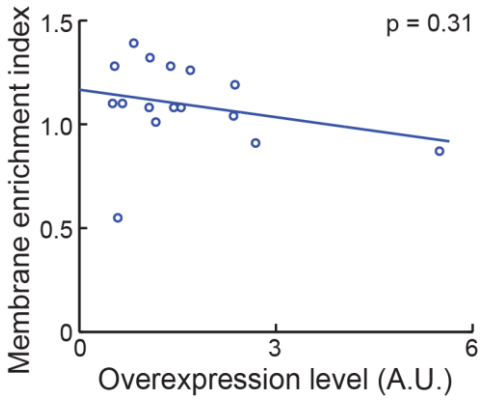

Fig. S2. Membrane enrichment indexes (MEI) of PKA-Cn are not dependent on the protein expression level.

The MEIs of PKA-Cn (same as those in Fig. 1c) did not correlate with the corresponding protein expression levels. The linear fit extrapolated to zero overexpression level at 1.17.

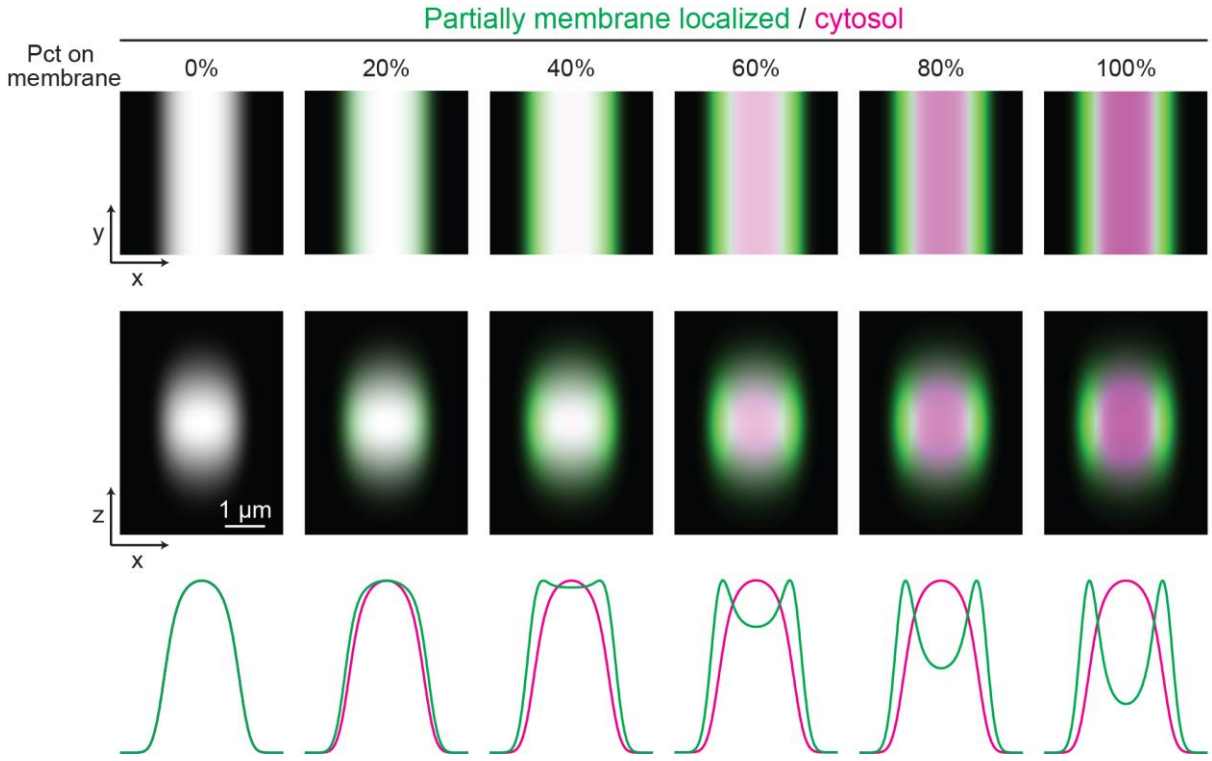

Fig. S3. Representative simulated images and traces of protein localization with different fractions on the membrane.

Proteins (green) with indicated percentage (pct) on the membrane of a model cylindrical neuronal dendrite (ϕ = 2.18 μm) along the y axis, with the remaining evenly distributed in the cytosol, are convoluted with a Gaussian simulated point spread function (PSF) with a lateral full-width-half-maximum (FWHM) size of 0.505 μm and an axial size of 1.78 μm. Both the x-y view (top) and the x-z view (middle) are shown and overlaid on the image of a fully cytosolic marker (magenta). Bottom panels show the normalized fluorescence intensity traces along the x axis at the z center.

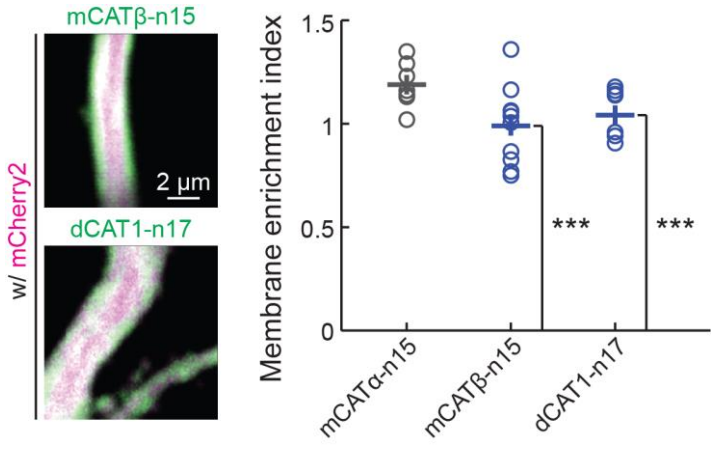

Fig. S4. The membrane affinity is evolutionally conserved across species and PKA-C isoforms.

Representative images (left) and the membrane enrichment indexes of the first 15 residues of mouse PKA-C α subunit (mCATα-n15, same as PKA-Cn15 in Fig. 1*C*), the first 15 residues of mouse PKA-C β subunit (mCATβ-n15), and the first 17 residues of *Drosophila* PKA-C CAT1 subunit (dCAT1-n17). See Fig. 1*A* for the exact sequences. n = 10 for mCATβ-n15, and 8 for dCAT1-n17.

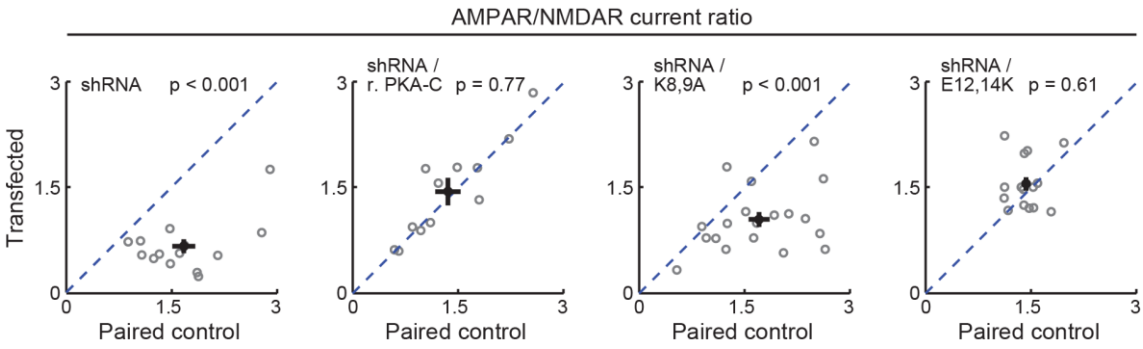

Fig. S5. AMPA/NMDA receptor current ratio.

Scatter plots of paired AMPA receptor to NMDA receptor current ratios from neighboring untransfected CA1 neurons paired with those transfected with shRNA against PKA-C and the indicated shRNA-resistant rescuing constructs. Statistical p values were tested using a sign test (MATLAB). From left to right, n = 12, 14, 19, and 15.