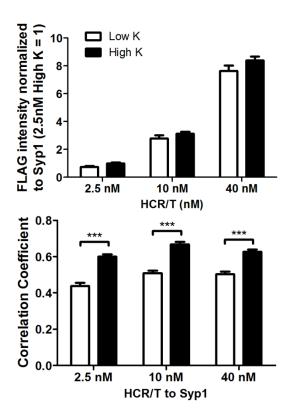


Supplementary Figure 1. HCR/T and HCR/A binding to neurons. 40 nM FLAG-HCR/T and FLAG-HCR/A were incubated with rat cortical neurons in low K buffer at 4°C for 60 min. Cells were washed, fixed and incubated with Alexa594-conjugated wheat germ agglutinin (WGA) for 15 min. Cells were then washed and fixed again. HCR bound to neurons was detected by immunofluorescence (IF) using mouse-α-FLAG antibody followed by alexa488-secondary antibody. **(A)** 40 nM FLAG-HCR/T and FLAG-HCR/A bound to rat cortical neurons under indicated binding buffer. Scale bar is 10 μm. **(B)** Quantified binding of FLAG-HCR/T and FLAG-HCR/A to rat cortical neurons using the intensity ratio between α-FLAG antibody and WGA staining. **(C)** Cell lysates were also collected and subjected to SDS-PAGE. Proteins were transferred to PVDF membranes and subjected to Western blotting, using α-3x-FLAG antibody (HCR) and α-actin antibody. The Western blot is shown.



Supplementary Figure 2. Dose-dependent association of HCR/T to neurons. 2.5, 10 or 40 nM FLAG-HCR/T was incubated with rat cortical neurons in low K buffer or high K buffer for 5 min at 37°C. Neurons were fixed and stained with α -FLAG and α -Syp. FLAG intensity was normalized to Syp1 signal (upper panel). Correlation coefficients (lower panel) were determined using ImageJ as described in methods. HCR/T/ α -syp ratios in low K buffer or high K buffer were not statistically different at the assayed concentrations.