SUPPLEMENTARY MATERIALS AND METHODS

Extracellular Slice Electrophysiology. Slices were kept at an ACSF interface in a humidified holding chamber for at least one hour prior to recordings. Slices were then transferred to a submersion-type recording chamber and superfused at a rate of 1.5 ml/min at room temperature (20-22°C). Concentric bipolar tungsten electrodes were used to electrically stimulate (100 µs duration at 0.05 Hz) Schaffer collateral afferents in the stratum radiatum (SC-CA1), or temporoammonic afferents in the stratum lacunosum-moleculare (TA-CA1). Under standard experimental conditions, ACSF-filled recording pipettes (3-5 MΩ) were placed in the stratum radiatum of the CA1 field to record field excitatory postsynaptic potentials (fEPSPs) evoked by Schaffer collateral stimulation. For dual electrophysiological recordings of SC- and TA-stimulation evoked fEPSPs, both SC and TA afferents were stimulated in an alternating fashion, and recording pipettes were placed in the stratum radiatum and the stratum lacunosum-moleculare of the CA1 field, respectively. Stimuli were set to 150% of threshold intensity, resulting in fEPSPs of 0.2-0.5 mV. Responses were amplified 1000×, filtered at 3 kHz, and digitized at 10 kHz. pClamp software was used to analyze the slope (mV/ms) of the initial 1.5-ms rising phase of the fEPSP. Only recordings exhibiting a stable presynaptic fiber volley across the experiment and <15% increase/variation in the fEPSP during baseline were included in the analyses. Modest changes in the fiber volley occurred as a result of nominally Mg^{2+}-free conditions, and were accounted for by normalizing the slope of the fEPSP to the fiber volley amplitude on a response-by-response basis. Test compounds were applied by bath application. Individual slope values plotted as a function of time reflect the mean of three consecutive sweeps (1 min). Paired pulse ratios with an interstimulus interval of 50 ms were generated at the end of the baseline recording, and again, during the last minute of the test compound superfusion; paired pulse values were calculated from the mean of five consecutive sweeps, second pulse (P2)/first pulse (P1). Change in paired pulse was calculated as P2-P1/P1x100.

Whole-cell Slice Electrophysiology. Slices were stored in an immersion ACSF holding chamber for at least one hour, at which time they were transferred to a submersion-type recording chamber and

superfused at a rate of 2 ml/min at room temperature (20-22°C). Recording pipettes (3.5-5 MΩ) were filled with a solution that contained (in mM): 130 Cs methanesulfonate, 10 CsCl; 2 $MgCl_2$, 10 EGTA, 20 CsOH, 10 HEPES (pH adjusted with CsOH to 7.3). mEPSCs were recorded using an LM-EPC7 amplifier (List Electronic, Darmstadt, Germany), filtered at 3 kHz, digitized at 10 kHz (Digidata 1332A, Molecular Devices Corp., Union City, CA), and acquired using the Clampex 9.2 software (Molecular Devices Corp.). Amplitude and frequency of mEPSCs were analyzed using the Mini Analysis software (v. 6.0.7, Synaptosoft Inc., Fort Lee, NJ). mEPSCs were identified with the amplitude threshold of 4-5 pA. The amplitude threshold for automated event detection was set at three times the standard deviation of the baseline noise. Events automatically detected by the software were visually inspected and manually rejected if they did not show a typical synaptic waveform. The detection threshold was fixed for all recordings obtained from any given cell. Recordings exhibiting a stable baseline and a current leak below 100 pA were included in the analyses. Individual data points plotted as a function of time reflect the mean amplitude or mean frequency of mEPSCs recorded in 5 min bins. Cumulative distribution plots of mEPSC amplitude and interevent interval were generated using a total of 250 events randomly selected from recordings obtained for the 5 min before and between 20 and 25 min after beginning of superfusion of the cells in each treatment group (VEH vs. (*2R,6R*)-HNK).

Chemicals. 6,7-dinitroquinoxaline-2,3(1H,4H)-dione (DNQX, 50 µM; Sigma-Aldrich, St. Louis, MO) was used to block the AMPAR-mediated component of the fEPSP, and 2-amino-5-phosphonopentanoic acid (AP5, 80 µM; Tocris) was used to block the NMDAR-mediated component of the fEPSP. The positive allosteric modulator of the AMPAR, phenyl-1,4-bis-alkylsulfonamide (CMPDA, 1 µM; Tocris) was used to potentiate AMPAR-mediated synaptic transmission. Recording ACSF for whole-cell mEPSCs contained the sodium channel blocker, tetrodotoxin (TTX, 0.3 µM; Enzo Life Sciences, Farmingdale, NY), and the NMDA-, $GABA_A$-, and $GABA_B$ receptor blockers, AP5 (80 µM; Sigma-Aldrich, St. Louis, MO), picrotoxin (100 µM; Tocris), and CGP52432 (2 µM; Tocris), respectively. Recording pipettes for whole-cell experiments contained the sodium channel blocker, QX-314 (Lidocaine *N*-ethyl bromide; Sigma-Aldrich, St. Louis, MO). In 80% of the cells, 6-cyano-7-

nitroquinoxaline-2,3-dione (CNQX, 10 µM; Sigma-Aldrich, St. Louis, MO) was bath applied, confirming that mEPSCs were AMPAR-mediated. All compounds were diluted to their final concentration in ACSF.

Experimental Design and Statistical Analysis. Data, which were found to be normally distributed (D'Agostino & Pearson) and homogenous (Brown-Forsythe), are presented as mean ± standard error of the mean. For extracellular slice electrophysiology experiments, statistical comparisons were made by comparing the mean baseline response to the mean response during the last 5 min of superfusion with the test compound. For whole-cell electrophysiology experiments, statistical comparisons were made by comparing the mean baseline results to the mean of results obtained during the first significant time-bin (15-20 min) of the superfusion of the slices with the test compounds. Within slice two-group comparisons were assessed with paired two-tailed Student's *t* tests, whereas between-slice two-group comparisons (e.g., whole-cell electrophysiology experiments) were assessed with unpaired Student's *t* tests. Cumulative distribution plots of inter-event intervals and amplitude were analyzed by the Kolmogorov-Smirnov test. Between group comparisons of three or more groups were assessed with one-way analysis of variance (ANOVA) followed by Holm-Šídák *post-hoc* comparisons. Two-way repeated measures ANOVA were used when drug condition (0.9% saline vehicle [VEH] vs. (*2R,6R*)-HNK) and pathway (SC-CA1 vs. TA-CA1; *repeated measure*), and when treatment (VEH vs. (*2R,6R*)-HNK) and time (*repeated measure*) were independent factors. If a significant interaction was detected, the Holm-Šídák *post-hoc* test was used for pairwise group comparisons. Planned comparisons were made once treatment effects were established at SC-CA1 synapses (e.g., SC-CA1 vs. TA-CA1 dual recording experiment). A four-parameter Hill function was used to derive the EC_{50} and IC_{50} from the concentration-response relationships, and linear regression was used to identify significant correlations between paired pulse ratios and fEPSP slopes.