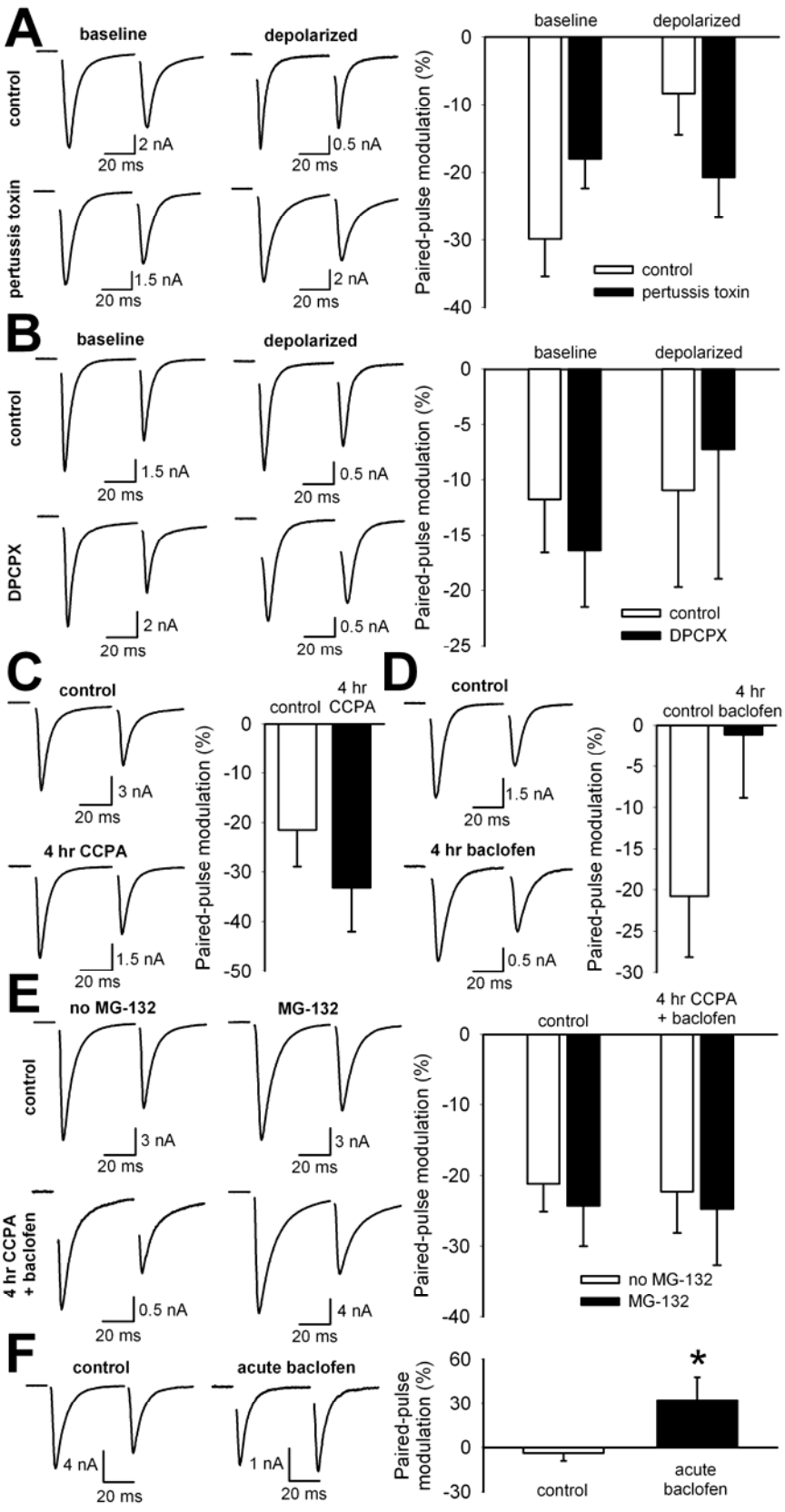
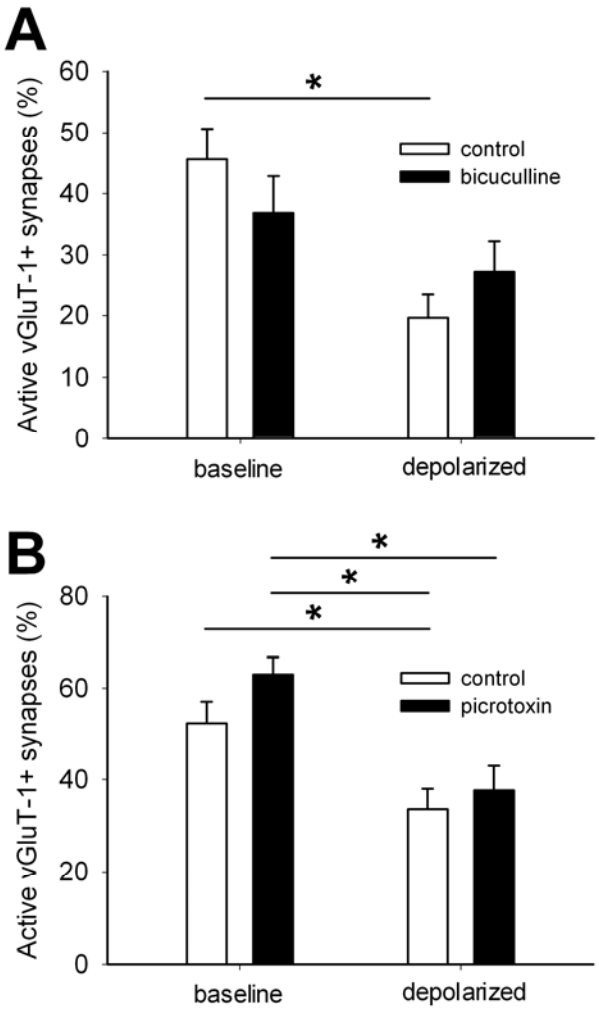
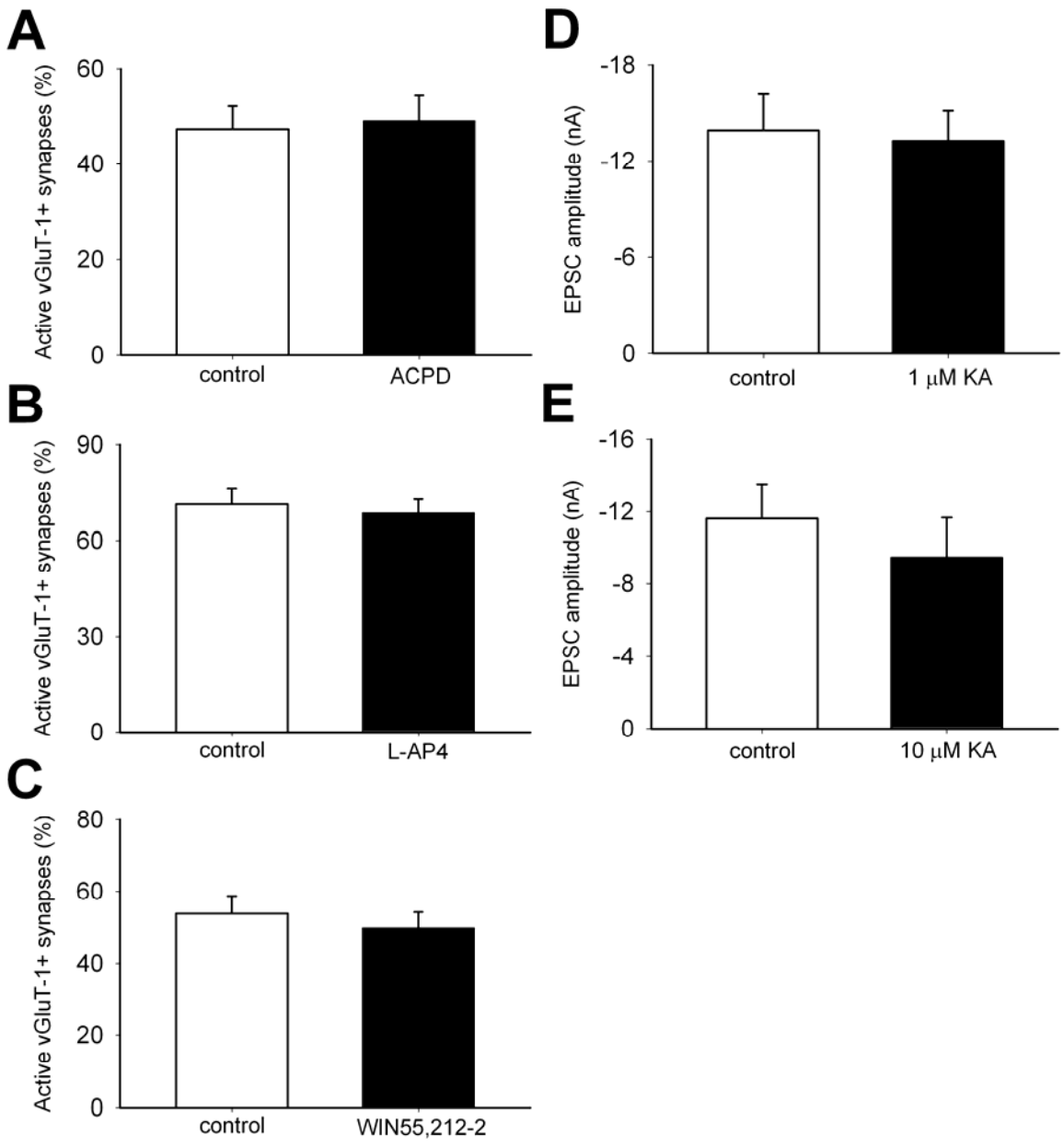

A
baseline
depolarized
control
pertussis toxin
2 nA
0.5 nA
1.5 nA
2 nA
20 ms
baseline
depolarized
Paired-pulse modulation (%)
control
pertussis toxin
B
DPCPX
1.5 nA
0.5 nA
2 nA
0.5 nA
control
DPCPX
C
control
4 hr CCPA
3 nA
1.5 nA
control
4 hr CCPA
D
4 hr baclofen
1.5 nA
0.5 nA
control
4 hr baclofen
E
no MG-132
MG-132
4 hr CCPA + baclofen
3 nA
0.5 nA
4 nA
control
4 hr CCPA + baclofen
no MG-132
MG-132
F
acute baclofen
4 nA
1 nA
*
control
acute baclofen

Supplemental Figure 1. Acute, but not prolonged, treatments alter paired-pulse modulation in excitatory autaptic neurons. **A.** Left panel: example action-potential evoked EPSCs with 50 ms interstimulus interval from autaptic neurons treated 24 hr with or without 500 ng/ml pertussis toxin before 4 hr co-application with 30 mM NaCl (baseline) or 30 mM KCl (depolarized). Right panel: summary of percentage change in EPSC amplitude during 50 ms paired-pulse stimulus in neurons treated as described in the left panel. p = 0.09-1.00 with Bonferroni correction for multiple comparisons (n = 13-14). **B.** Left panel: example action-potential evoked EPSCs with 50 ms interstimulus interval from autaptic neurons treated 4 hr with 30 mM NaCl (baseline) or 30 mM KCl (depolarized) in the presence or absence of 200 nM DPCPX. Right panel: summary of percentage change in EPSC amplitude during 50 ms paired-pulse stimulus in neurons treated as described in the left panel. p = 0.59-1.00 with Bonferroni correction for multiple comparisons (n = 9). **C.** Left panel: example action-potential evoked EPSCs with 50 ms interstimulus interval from autaptic neurons treated either acutely (< 1 min; control) or for 4 hr with 10 nM CCPA. Right panel: summary of percentage change in EPSC amplitude during 50 ms paired-pulse stimulus in neurons treated as described in the left panel. p = 0.79 (n = 9-10). **D.** Left panel: example action-potential evoked EPSCs with 50 ms interstimulus interval from autaptic neurons treated either acutely (< 1 min; control) or for 4 hr with 50 μM baclofen. Right panel: summary of percentage change in EPSC amplitude during 50 ms paired-pulse stimulus in neurons treated as described in the left panel. p = 0.08 (n = 9). **E.** Left panel: example action-potential evoked EPSCs with 50 ms interstimulus interval from autaptic neurons treated acutely (< 1 min; control) or for 4 hr with 10 nM CCPA and 50 μM baclofen with or without 3 μM MG-132. MG-132 was added 30 min prior to the start of (and remained during) the 4 hr treatment. Right panel: summary of percentage change in EPSC amplitude during 50 ms paired-pulse stimulus in neurons treated as described in the left panel. p = 0.66-0.97 without correction for multiple comparisons (n = 14-15). **F.** Left panel: example action-potential evoked EPSCs with 50 ms interstimulus interval from autaptic neurons treated acutely with locally perfused saline control or 10 μM baclofen. Right panel: summary of percentage change in EPSC amplitude during 50 ms paired-pulse stimulus in neurons treated as described in the left panel. *p = 0.04 (n = 12).



Supplemental Figure 2. Blocking $GABA_A$ receptors does not prevent depolarization-induced silencing. **A.** Summary of experiments measuring vGluT-1/FM1-43FX correspondence in neurons treated 4 hr with 30 mM NaCl (baseline) or 30 mM KCl (depolarized) in the presence or absence of 50 μM bicuculline, a $GABA_A$ receptor antagonist. *$p < 0.05$ with Bonferroni correction for multiple comparisons ($n = 25$ fields from 5 independent experiments). **B.** Summary of experiments measuring vGluT-1/FM1-43FX correspondence in neurons treated 4 hr with 30 mM NaCl (baseline) or 30 mM KCl (depolarized) in the presence or absence of 100 μM picrotoxin, another $GABA_A$ receptor antagonist. *$p < 0.05$ with Bonferroni correction for multiple comparisons ($n = 30$ fields from 6 independent experiments).



Supplemental Figure 3. mGluR, CB1, and kainate receptor agonists do not induce silencing. **A.** Summary of experiments measuring vGluT-1/FM1-43FX correspondence in neurons treated 4 hr with or without 200 μM ACPD (mGluR agonist; $n = 30$ fields from 6 coverslips; $p = 0.81$). **B.** Summary of experiments measuring vGluT-1/FM1-43FX correspondence in neurons treated 4 hr with or without 10 μM L-AP4 (mGluR agonist; $n = 15$ fields from 3 coverslips; $p = 0.69$). **C.** Summary of experiments measuring vGluT-1/FM1-43FX correspondence in neurons treated 4 hr with or without 1 μM WIN55,212-2 (CB1 agonist; $n = 40$ fields from 8 coverslips; $p = 0.54$). **D.** Summary of experiments measuring EPSC amplitude in neurons treated 4 hr with or without 1 μM kainic acid (KA; kainate receptor agonist; $n = 12$ neurons; $p = 0.82$). **E.** Summary of experiments measuring EPSC amplitude in neurons treated 4 hr with or without 10 μM kainic acid ($n = 11$ neurons; $p = 0.47$).