SUPPLEMENTARY INFORMATION

Differential control of presynaptic efficacy by postsynaptic N-cadherin and β-catenin

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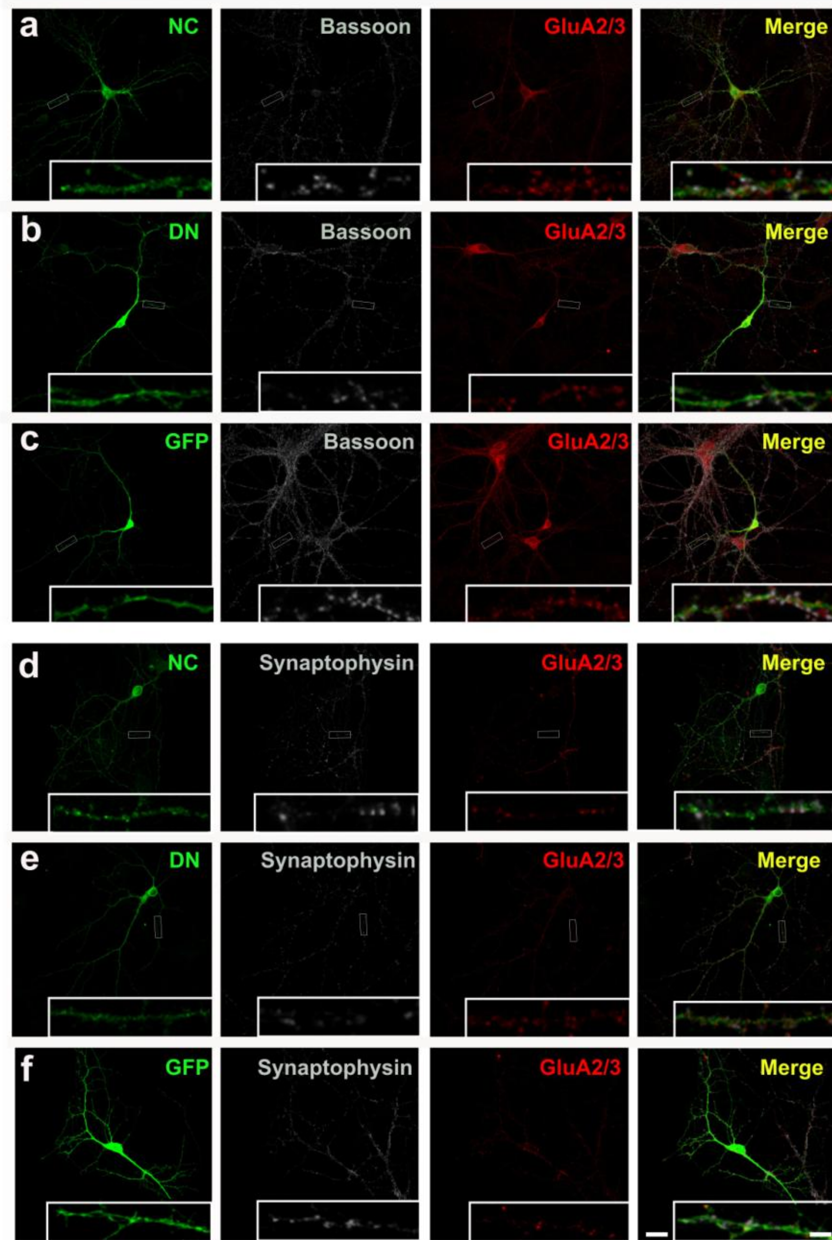
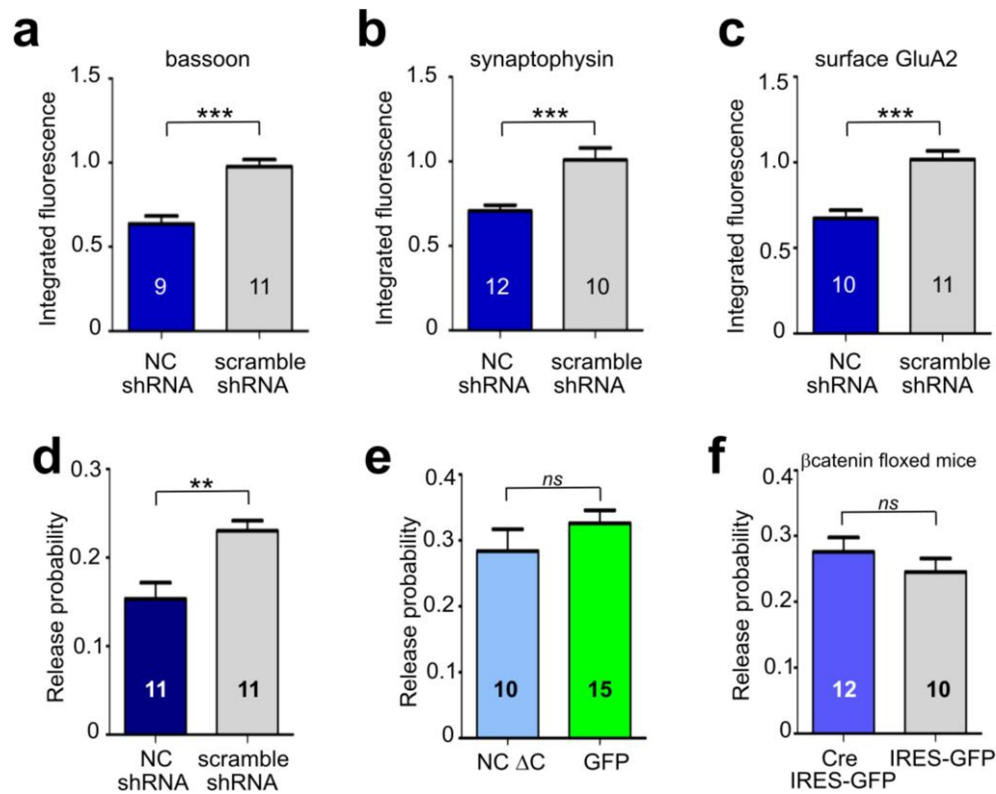
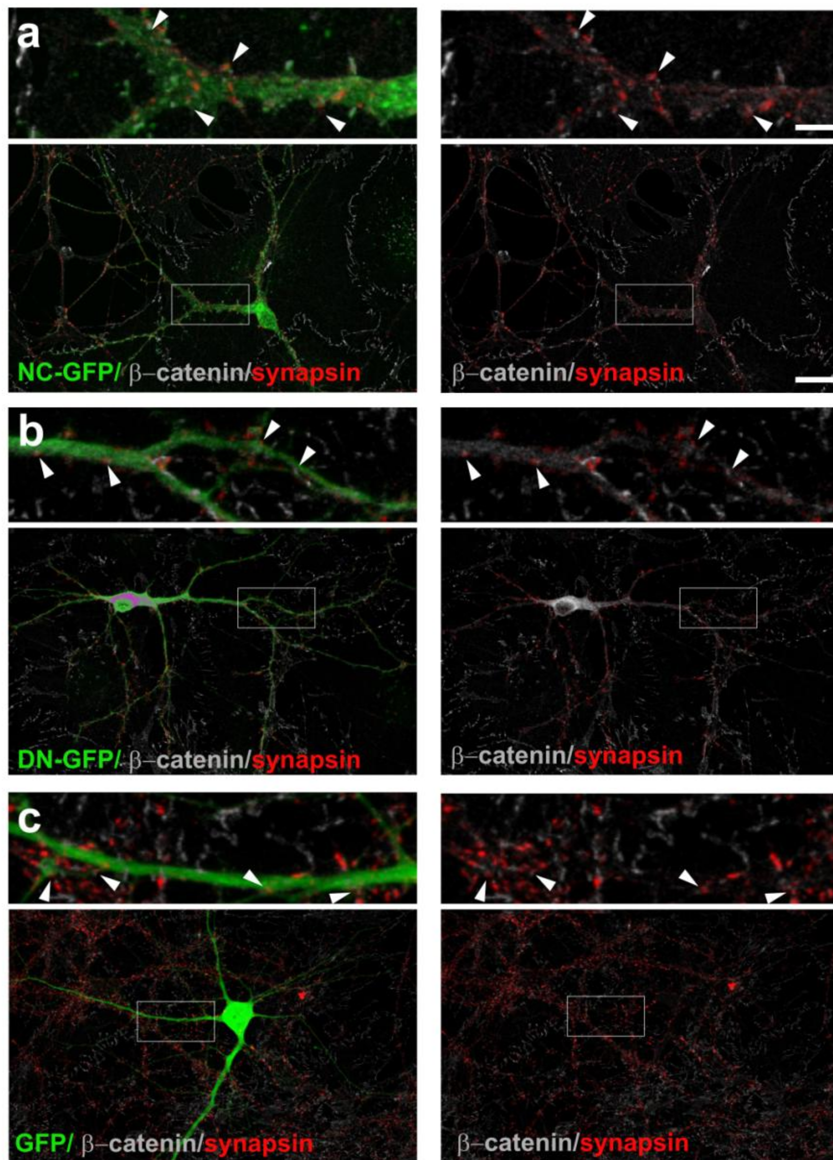
Supplementary Figures 1 - 6



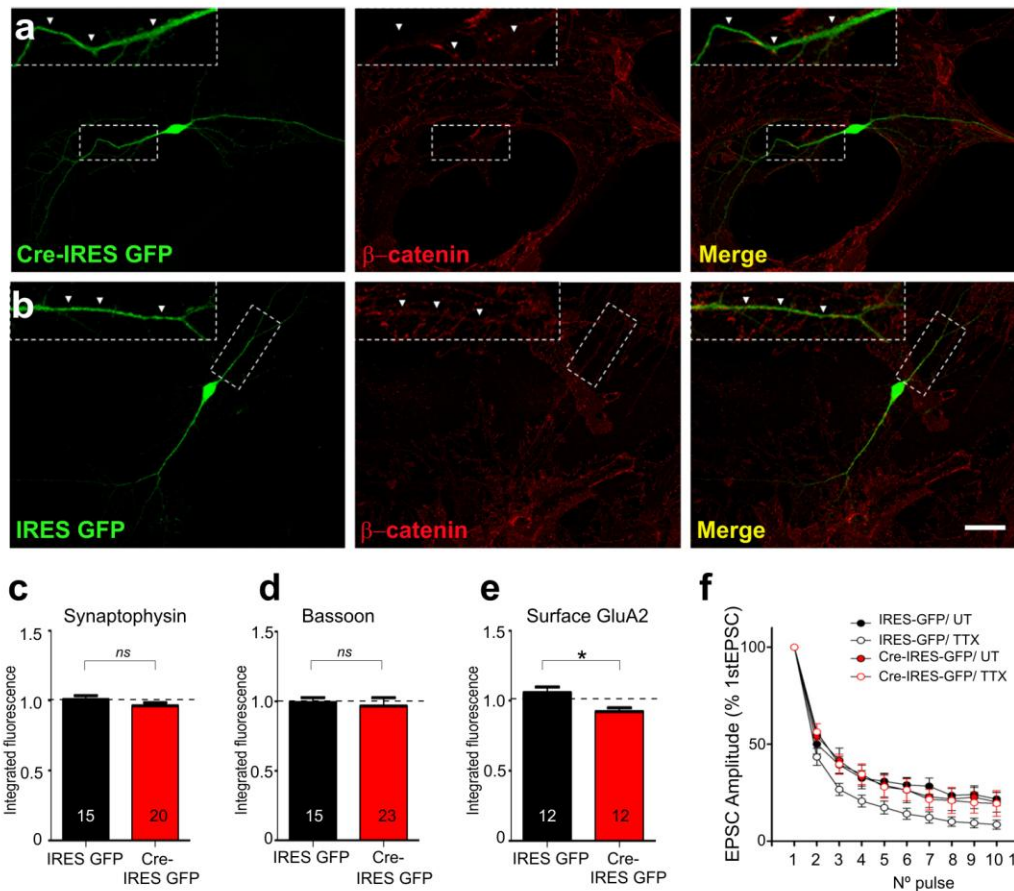
Supplementary Figure 1. Postsynaptic expression of DN-NCad reduces expression of presynaptic proteins. Representative images of hippocampal neurons transfected with WT-NCad **(a, d)**, DN-NCad **(b, e)** and GFP **(c, f)** and double labeled for bassoon or synaptophysin and GluA2/3. Scale bars, 20 µm; inset 6 µm.



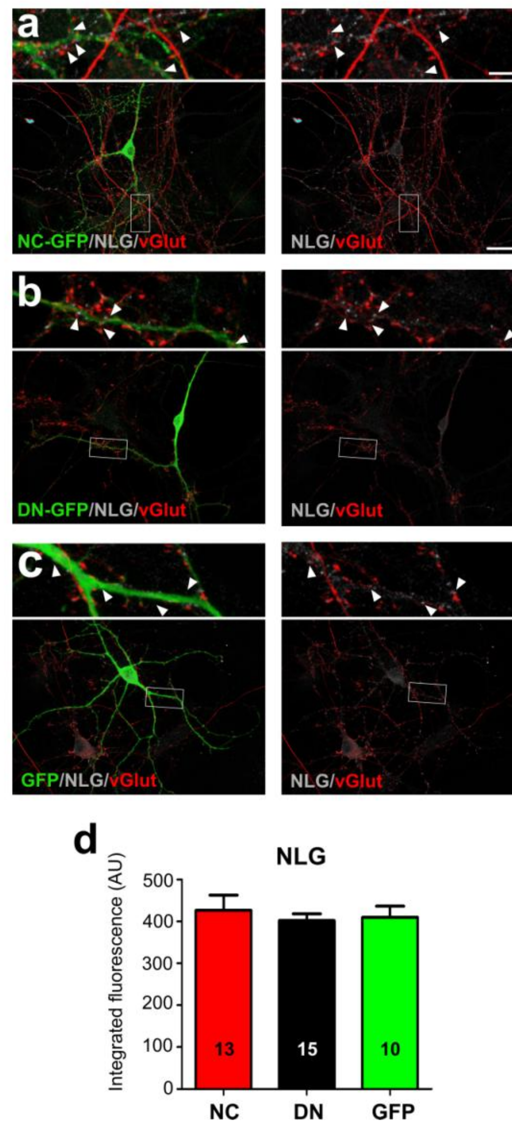
Supplementary Figure 2. Postsynaptic N-cadherin knock-down decreases the levels of presynaptic proteins and surface GluA2. a-c, Summary of integrated immunofluorescence puncta intensity in cells postsynaptically expressing NC shRNA and control scramble. Synaptic puncta were identified by double labeling for bassoon or synaptophysin and GluA2/3, or for synapsin and surface GluA2. Bars show mean ± SEM for each group relative to untransfected neighboring neurons in the same field of view. **d-f,** Summary of p_r at synapses formed onto dendrites expressing a N-cadherin shRNA or control scrambled shRNA (d), a mutant N-cadherin construct (NC-ΔC) or GFP (e), and in β-catenin floxed hippocampal neurons transfected with Cre-IRES-GFP to knock down β-catenin or control IRES-GFP (f). Bars show mean ± SEM. The number of neurons used is shown in bars; ns denotes no statistically significant difference, **P<0.01, ***P<0.001; two tailed student's *t*-test.



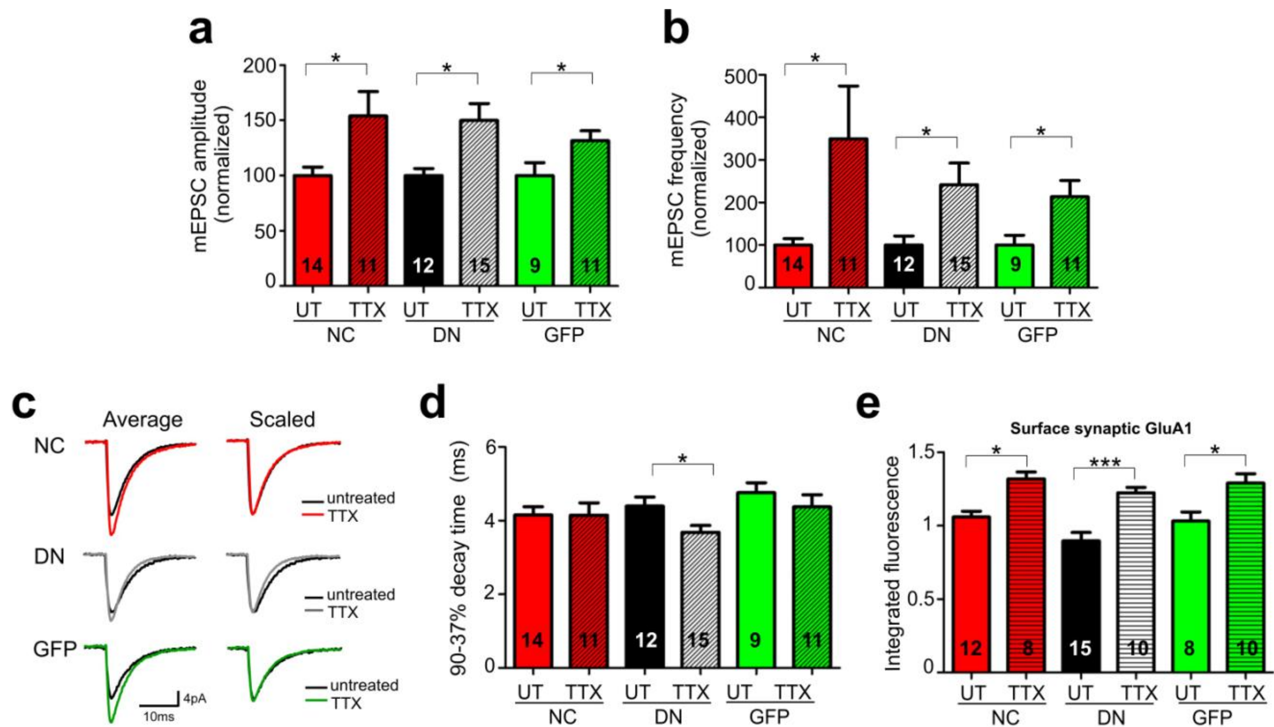
Supplementary Figure 3. Localization of β-catenin in neurons expressing WT-NCad, DN-NCad or control GFP. Hippocampal neurons postsynaptically expressing WT-NCad (a), DN-Ncad (b) or GFP alone (c) show a similar distribution of β-catenin. Cultures were double labeled for β-catenin and synapsin. Arrowheads show synaptic β-catenin apposed to synapsin. Scale bar: 50 µm; inset 5 µm.



Supplementary Figure 4. Postsynaptic β-catenin knock-down prevents the adjustment of p_r after activity block. a, b, Neurons from β-catenin floxed mice expressing Cre-IRES-GFP show reduced levels of β-catenin compared to control cells. **c-d,** Summary of puncta intensity of indicated pre- and postsynaptic proteins. Bars show mean ± SEM relative to non-transfected neighboring neurons in the same field of view. The number of neurons used is shown in bars; *P<0.05; ns*,* no statistically significant difference; two tailed student's *t*-test . Scale bar, 50 µm. **f**, Averaged EPSC amplitudes of neurons expressing Cre-IRES GFP treated with (n= 8 cell pairs) or without TTX (n=9 cell pairs) or control IRES GFP treated with (n=9 cell pairs) or without TTX (n=8 cell pairs). Two tailed student's *t*-test, P>0.05.



Supplementary Figure 5. Disruption of postsynaptic N-cadherin does not affect the levels of neuroligin at excitatory synapses. a-c, Hippocampal synapses postsynaptically expressing WT-NCad (a), DN-Ncad (b) or GFP alone (c) show a similar level of synaptic neuroligin (NLG) immunofluorescence signal. Excitatory synapses are identified by double labeling for vGlut1 (arrowheads). **d**, Summary of NLG associated integrated fluorescence intensity. Bars show mean ± SEM for each group. The number of neurons is indicated in bars. Scale bar: 50 µm; inset 6 µm.



Supplementary Figure 6. Disruption of postsynaptic N-cadherin activity alters the subunit composition of AMPARs. a-b, Summary of the effect of chronic activity block with TTX (1 µM, 36 h) on mEPSC amplitude (a) and frequency (b) corresponding to Figure 7b, c, showing the data normalized to untreated control. **c,** Effect of TTX treatment on mEPSC waveform of synapses expressing WT-NCad, DN-NCad or control GFP. Representative averaged traces of mEPSCs illustrate a faster decay following TTX treatment in DN-NCad neurons (c_i). Summary of mean mEPSC decay times (c_{ii}); two tailed student's *t*-test, P*<0.05. **d,** Summary of integrated puncta intensity of surface synaptic GluA1 labeling in cells postsynaptically expressing WT-NCad, DN-NCad or GFP. Synaptic GluA1 was identified by double labeling for synapsin. Bars show mean ± SEM relative to control neighboring untransfected neurons without TTX treatments. The number of neurons used is shown in bars; *P<0.05, ***P<0.001. One way ANOVA followed by Tukey's test.