Supplementary figure legends

Supplementary Figure 1: (a) pH sensitivities of receptors incorporating GluN1wt and different chimeric subunits exchanging the NTDs and/or the NTD-ABD linkers of GluN2A and GluN2B subunits. The pH IC_{50}s are from top to bottom: 7.41 ± 0.003 (n = 15), 8.1 ± 0.1 (n = 5), 7.14 ± 0.03 (n = 4), 7.40 ± 0.02 (n = 4), 7.26 ± 0.02 (n = 4), 7.02 ± 0.02 (n = 4), 7.18 ± 0.02 (n = 4), 7.26 ± 0.03 (n = 4) and 6.91 ± 0.02 (n = 11). **(b)** Spermine dose-response curve of GluN1 wt/ GluN2B-2A(L) receptors. The spermine EC_{50}, maximal potentiation and Hill coefficient are, respectively: 195 ± 35 μM, 9.0 ± 0.7 and 1.3 ± 0.1 (n = 4). The dashed curve represents the spermine dose-response curve for GluN1 wt/GluN2B wt receptors (replotted from Fig. 2c).

Supplementary Figure 2: Spermine-induced potentiations of different mutant receptors composed of a GluN2B subunit containing either a cysteine or alanine point mutation and either (**a**) GluN1wt, (**b**) GluN1-E181C, (**c**) GluN1-E185C, (**d**) GluN1-E186C or (**e**) GluN1-E188C. Spermine was applied at 200 μM and the extracellular pH was 6.5. The lower and upper dashed lines indicate the values obtained for wild-type GluN1/GluN2A receptors ($I_{spermine}/I_0$ = 0.90 ± 0.04, n = 17), and GluN1/GluN2B receptors ($I_{spermine}/I_0$ = 8.0 ± 1.6, n = 16), respectively.

Supplementary Figure 3: Electrostatic potential surfaces of the GluN1 (pdb: 3Q41, Farina et al, 2011), GluN2B (pdb: 3JPW, Karakas et al, 2009) and GluN2A (homology model from Stroebel et al, 2011) NTDs viewed from their putative dimerization face. Dashed circles indicate the location of the acidic residues previously shown to influence spermine sensitivity.

Supplementary Figure 4: (**a**) pH sensitivities and (**b**) estimations of maximal channel activity (Po) of receptors incorporating GluN1wt and different GluN2A/GluN2B chimeric subunits. (**a**) The pH IC_{50} values are (from top to bottom): 7.41 ± 0.003 (n = 15), 7.35 ± 0.01 (n = 3), 7.33 ± 0.01 (n = 3), 7.03 ± 0.02 (n = 5), 7.08 ± 0.01 (n = 4), 7.19 ± 0.02 (n = 5) and 6.91 ± 0.02 (n = 11). (**b**) MK-801 inhibition kinetics. Onset time constants (τ_{on}) were normalized to the value obtained for wild-type GluN1/GluN2B receptors. Values of are (from

top to bottom): 1.0 ± 0.1 (n = 86), 0.46 ± 0.06 (n = 7), 0.52 ± 0.07 (n = 11), 0.55 ± 0.07 (n = 7), 0.42 ± 0.06 (n = 9), 0.47 ± 0.08 (n = 10) and 0.34 ± 0.08 (n = 26). The left and right dashed lines indicate the values obtained for wild-type GluN1/GluN2A and GluN1/GluN2B receptors, respectively.

Supplementary Figure 5: Spermine-induced potentiations of mutant receptors composed of a GluN2B subunit containing either a single or double arginine or lysine point mutation and either (**a**) GluN1wt, (**b**) GluN1-E181R or (**c**) GluN1-E185R. Spermine was applied at 200 μM and the extracellular pH was 6.5. The lower and upper dashed lines indicate the values obtained for wild-type GluN1/GluN2A and GluN1/GluN2B receptors, respectively. Each bar represents the mean value from 3-13 different cells.

Supplementary Figure 6: MK-801 inhibition kinetics of mutant receptors composed of a GluN2B subunit containing either a single or double arginine or lysine point mutation and either (a) GluN1wt, (b) GluN1-E181R or (c) GluN1-E185R. Onset time constants (τ_{on}) were normalized to the value obtained for wild-type GluN1/GluN2B receptors. The lower and upper dashed lines indicate the values obtained for wild-type GluN1/GluN2A and GluN1/GluN2B receptors, respectively. The numbers in bold indicate the number of charge inversions (i). Each bar represents the mean value from 3-8 different cells (except for GluN1wt/GluN2B-E200R-E201R, n=1).

Supplementary references

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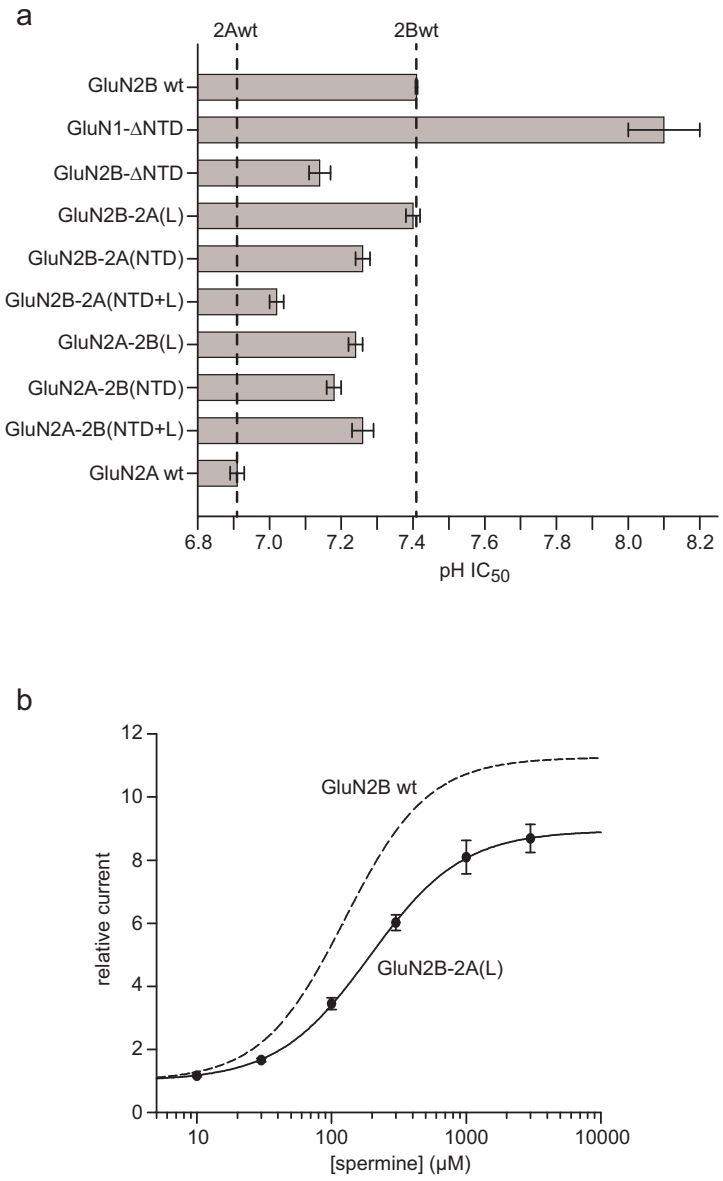
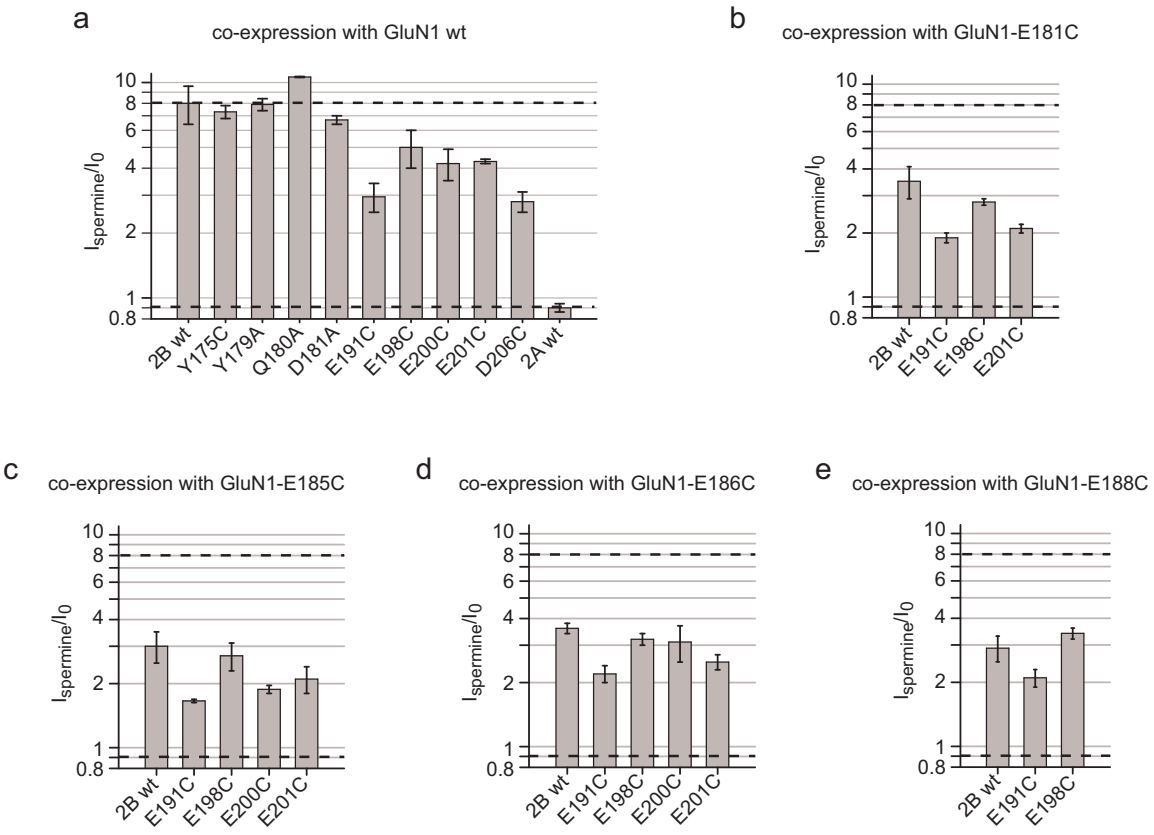
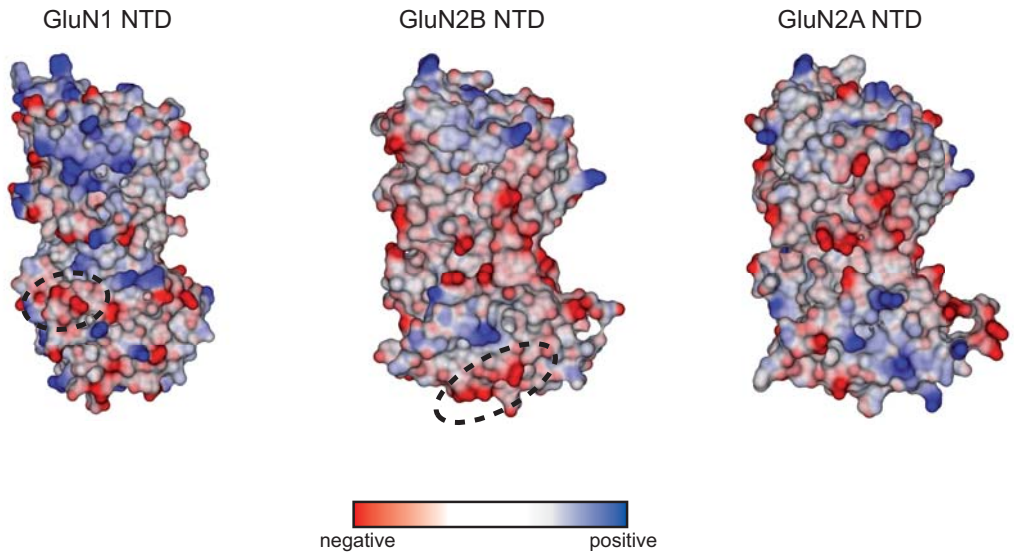


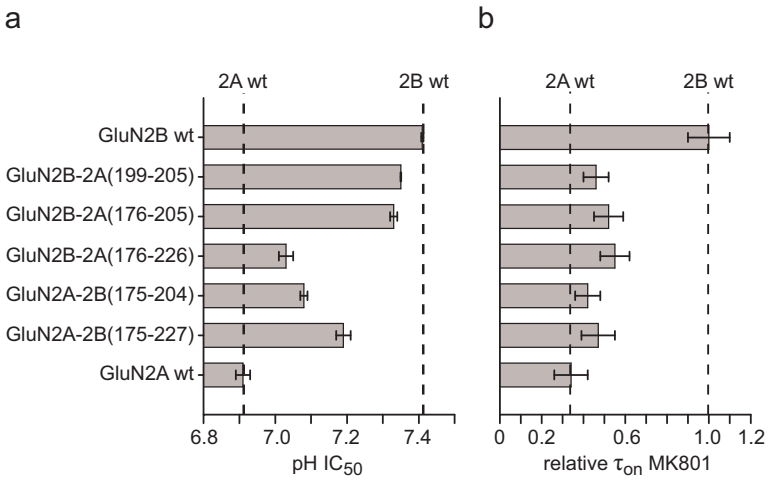
Supplementary Figure 1



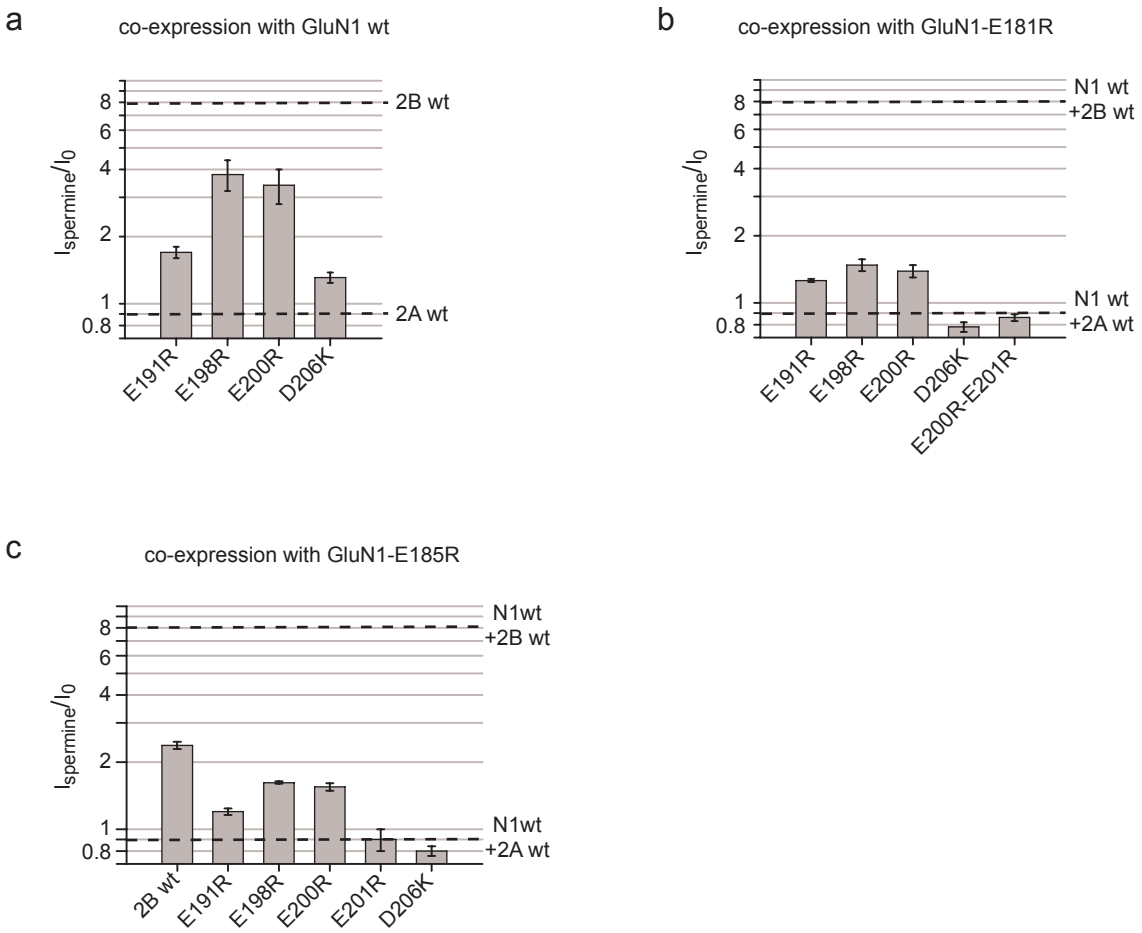
Supplementary Figure 2



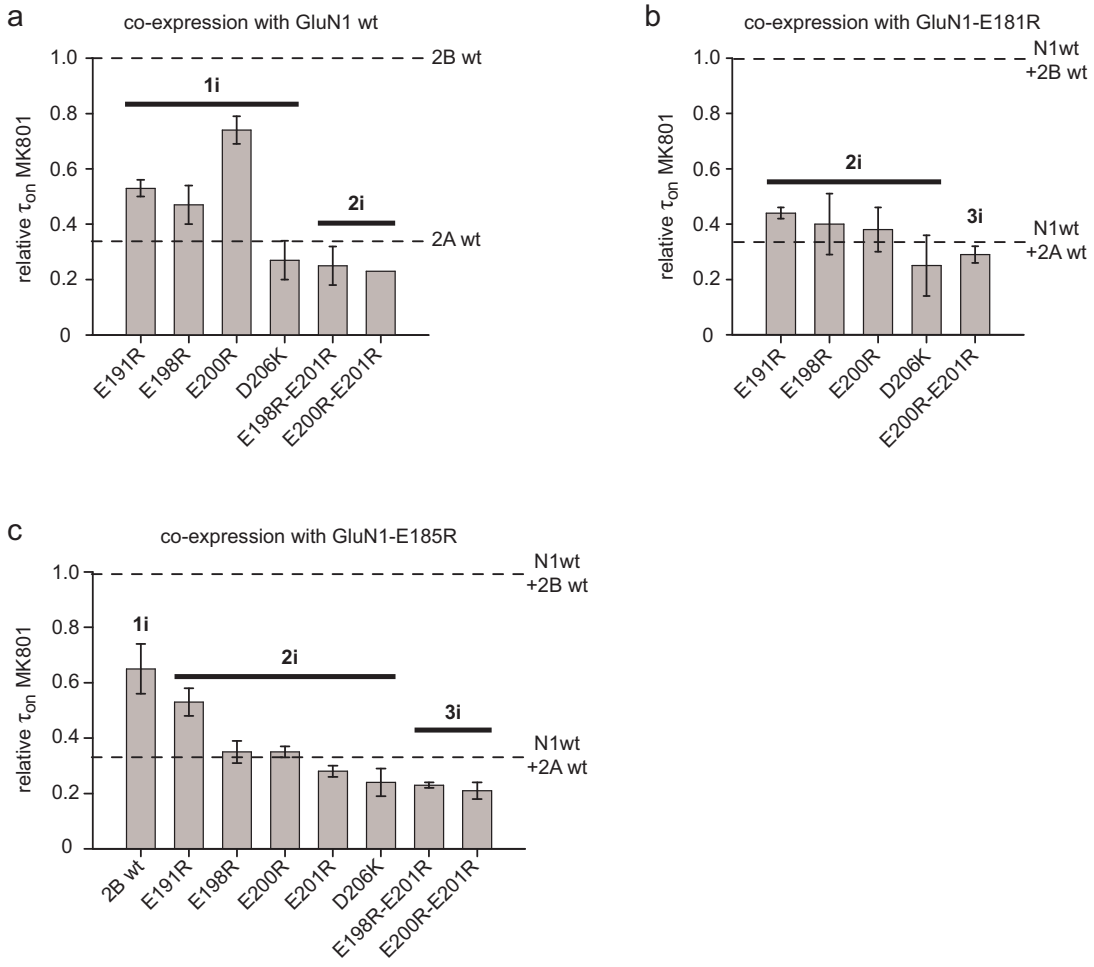
Supplementary Figure 3



Supplementary Figure 4



Supplementary Figure 5



Supplementary Figure 6