

Figure S5. Cortical neurons were transfected with plasmids expressing sh-Mfn2 or sh-sc (A) or (B) Mfn1 or control (globin). After 48 hours neurons were treated with NMDA (15 μ M in (A) or 30 μ M in (B)) for 6 hours. Neuronal death was analyzed (n=6 in (A), n=8 in (B)). *p<0.05, two-tailed T-test. C) NMDA (15 μ M) application for the indicated times to neurons transduced with AAV producing sh-Mfn2 or control sh-sc. NAD⁺ and NADH levels were analyzed independently and the ratio calculated (n= 4). Not significant differences found, two-tailed T-test. D) Cell death analysis of neurons expressing sh-Mfn2 and treated with NMDA (15 μ M) for 6 hours after 45 minutes pretreatment with CsA (1 μ M). (n= 5). *p<0.05, two-tailed T-test. E) Representative western blot showing spectrin full length (Spectrin-FL) and spectrin cleaved (spectrin-clev.) in neurons expressing sh-sc or sh-Mfn2 after treatment with 15 μ M NMDA for the indicated times. (n= 3). Analysis of the mitochondrial morphology of neurons expressing siCT or siBAX (F) or control plasmid (globin) or Bcl-xL (G) and treated with 30 μ M NMDA for 1 hour followed to washout and recovery for 3 hours. (n= 6). *p<0.05, two-tailed T-test.