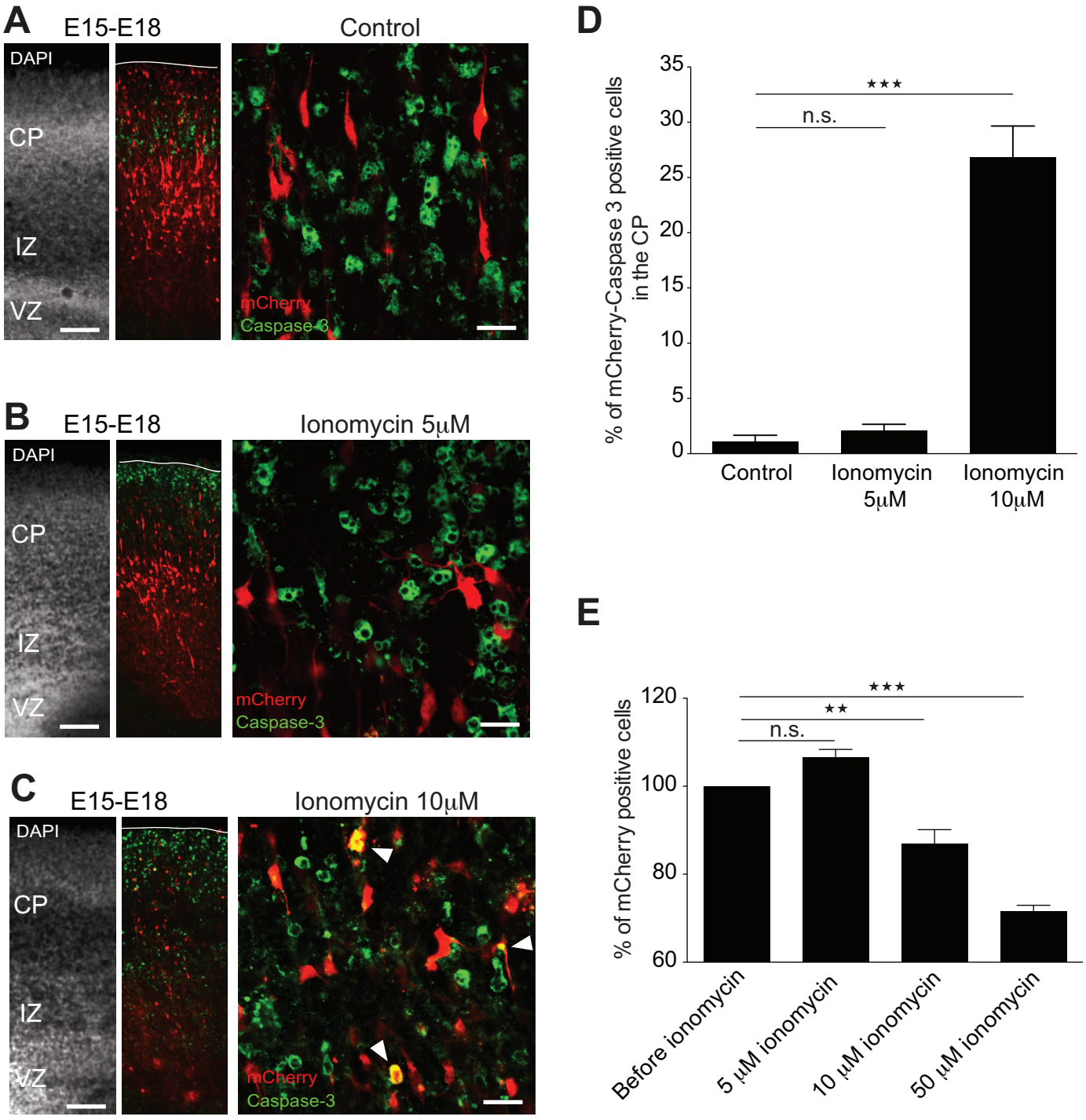
A
E15-E18
Control
DAPI
CP
IZ
VZ
mCherry
Caspase-3
B
E15-E18
Ionomycin 5μM
DAPI
CP
IZ
VZ
mCherry
Caspase-3
C
E15-E18
Ionomycin 10μM
DAPI
CP
IZ
VZ
mCherry
Caspase-3
D
% of mCherry-Caspase 3 positive cells in the CP
35
30
25
20
15
10
5
0
★★★
n.s.
Control
Ionomycin 5μM
Ionomycin 10μM
E
% of mCherry positive cells
120
100
80
60
★★★
★★
n.s.
Before ionomycin
5 μM ionomycin
10 μM ionomycin
50 μM ionomycin

Figure S4. Ionomycin has a dose-response effect in developing cortical neurons. (**A**) In control slices (*in utero* electroporated at E15, brain slices prepared at E18 and incubated 24 hr) low percentage (1.09 ± 0.57%) of mCherry-transfected cell in the CP express caspase-3. (**B**) Ionomycin 5 μM does not change the expression of caspase-3 in mCherry-transfected cell in the CP compared with control slices (2.12 ± 0.53%; *in utero* electroporated at E15, brain slices prepared at E18 and incubated 24 hr). (**C**) Ionomycin 10 μM increases significantly the expression of caspase-3 in mCherry-transfected cell in the CP compare with control slices (26.80 ± 2.85%; *in utero* electroporated at E15, brain slices prepared at E18 and incubated 24 hr). (**D**) Quantification from (A-C) showing percentage of mCherry-transfected cells positive to caspase-3 in the CP upon ionomycin treatment (Control = 1108 cells from fourteen slices from five brains from three litters; 5 μM = 1320 cells from twenty one slices from eight brains from three litters; 10 μM = 637 cells from twelve slices from four brains from three litters $p<0.0001$ by one-way ANOVA, posthoc Dunnett test ***$p<0.001$; values are mean ± s.e.m.). (**E**) Dose-response effect of ionomycin in cell number of mCherry transfected cells in the CP. Cortical slices were prepared for time-lapse analysis at E18 (*in utero* electroporated at E15). Time-lapse analysis was performed before (1 hr) and after ionomycin treatment (12 hr of ionomycin treatment). 50μM (71.48 ± 1.44%) and 10 μM (86.90 ± 3.23%) decreased significantly the number of mCherry transfected cells in the cortical plate after 12 hr imaging. 5μM did not decrease the number of mCherry-transfected cells in the CP (106.55 ± 1.82%; 5μM (before ionomycin) = 174 cells from four slices; 10μM (before ionomycin) = 180 cells from four slices; 50μM (before ionomycin) = 222 cells from four slices; $p<0.0001$ by one-way ANOVA, posthoc Dunnett test **$p<0.01$, ***$p<0.001$; values are mean ± s.e.m.). Scale bar: 200 μm (left panel) and 20 μm (right panel).