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Supplemental Information

Enhancing Electrotransfection Efficiency through Improvement in Nuclear Entry of Plasmid DNA

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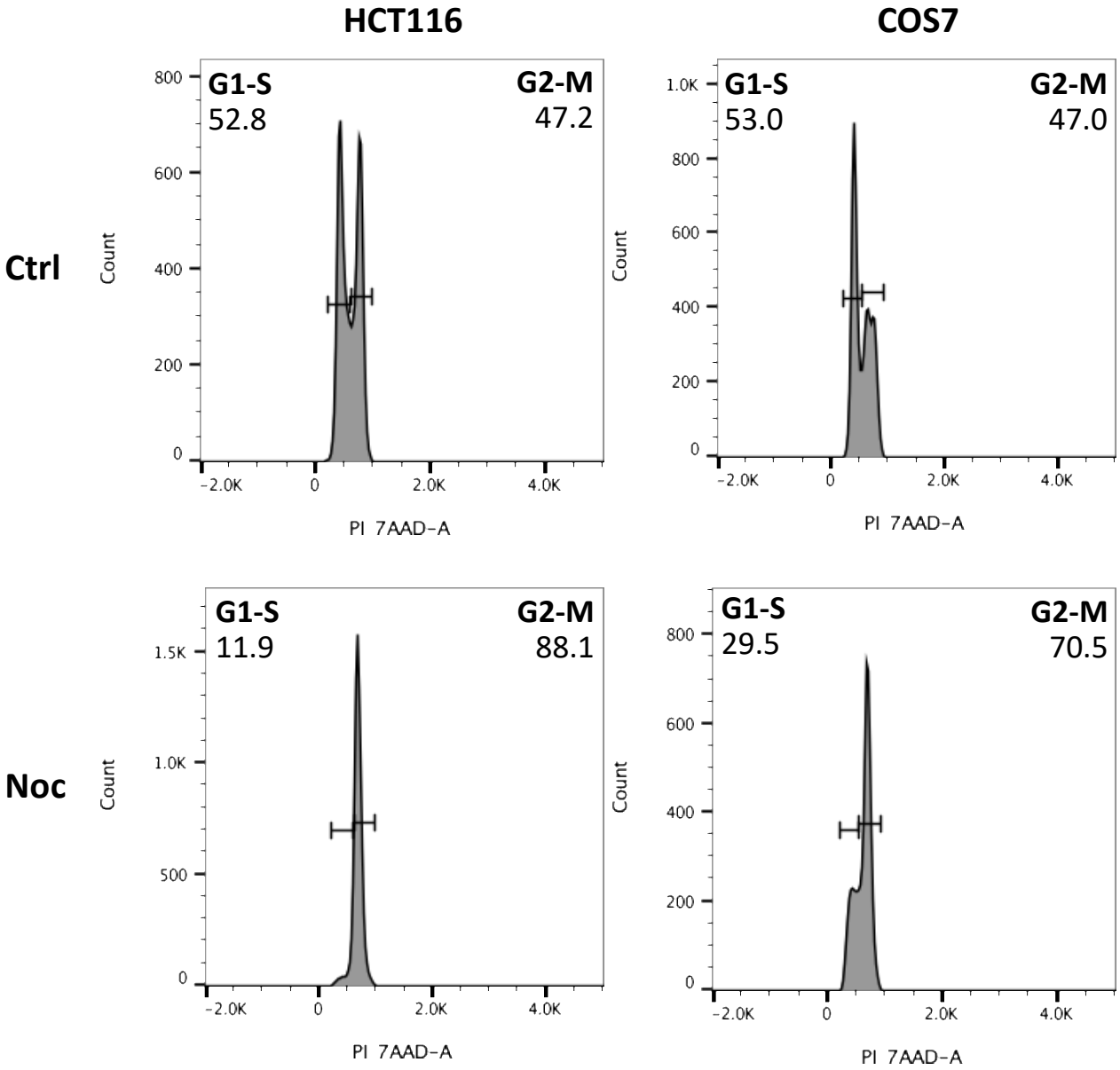
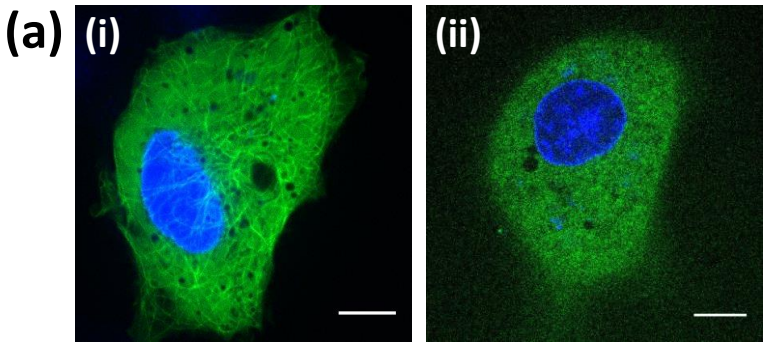
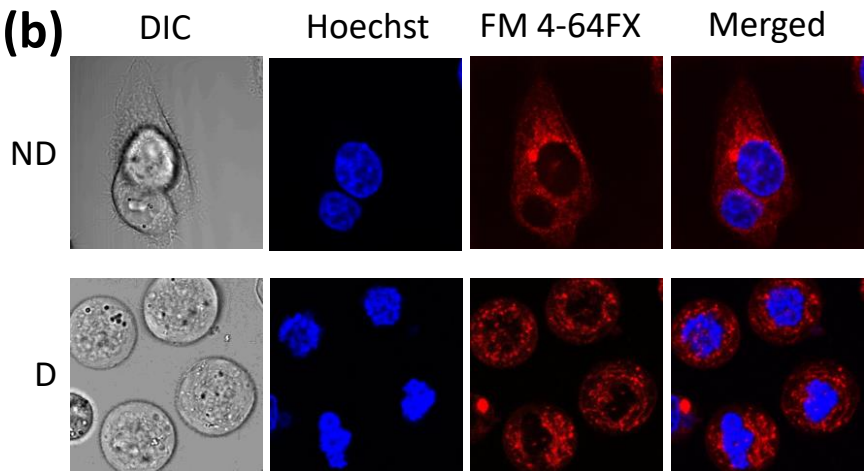
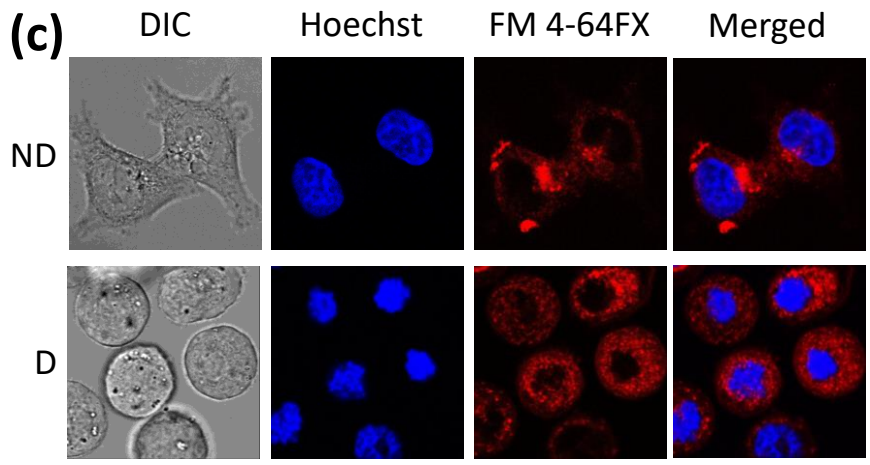


Figure S1. Cell cycle analysis for HCT116 and COS7 cells. The cells were treated with either nocodazole (Noc) or DMSO (Ctrl) for 16 hours. Cell populations in different phases were detected by flow cytometry after they were fixed and stained with propidium iodide (PI).


(a)
(i)
(ii)


(b)
DIC
Hoechst
FM 4-64FX
Merged
ND
D


(c)
DIC
Hoechst
FM 4-64FX
Merged
ND
D

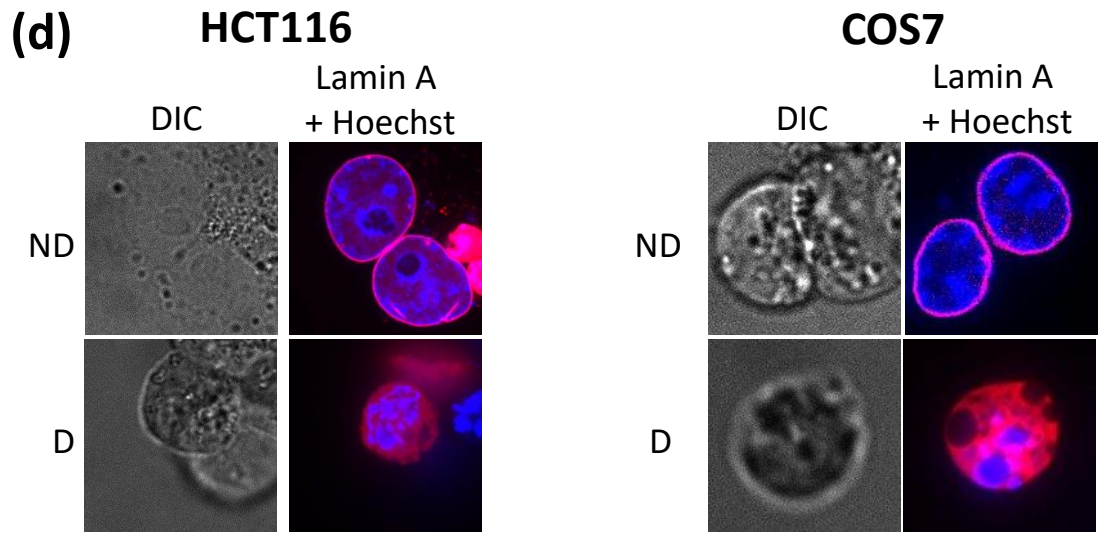


Figure S2. Microtubule depolymerization and nuclear envelope breakdown. In all panels, the images were acquired with a confocal microscopy after cells were treated with control vehicle or nocodazole (100 ng/ml) for 16 hours. Cells were at different phases in the control group, but synchronized at the G2-M phase in the nocodazole-treated group. To show that the nuclear envelope broke down (NEBD) in the M phase, we selected non-dividing (ND) cells from the control group and dividing (D) cells from the treated group. The images in the figure show the differences in morphology between the two cell states. The nuclei of cells were stained with Hoechst 33342 dye (blue). **(a)** Confocal images of HCT116 cells expressing a fusion protein of GFP-alpha-tubulin after nocodazole treatment (scale bar: 10 μm). (i) Control cells had fiber-like structures of microtubules, whereas (ii) nocodazole-treated cells had a diffuse pattern of alpha-tubulin, due to microtubule depolymerization. **(b, c)** The nuclear envelope was outlined by staining membranous structures with FM4-64FX dye (red) for 30 min either (b) after or (c) before nocodazole treatment of HCT116 cells. The cell nuclei stained with Hoechst dye were larger and had smooth surface in non-dividing control cells, but were smaller and contained many aggregates in dividing cells from the nocodazole-treated group. No apparent differences were observed in distributions of FM4-64FX between Panels (b) and (c). **(d)** The nuclear envelope was outlined by expressing a fusion protein of mCherry-Lamin A in HCT116 and COS7 cells. Non-dividing control cells displayed Lamin A around the Hoechst dye-stained nuclei. However, Lamin A in dividing cells from the nocodazole treated group showed a diffuse pattern ~~of the Hoechst dye~~.