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Supplemental Information

Light-Triggerable Liposomes for Enhanced Endolysosomal Escape and Gene Silencing in PC12 Cells

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Supplemental Figures

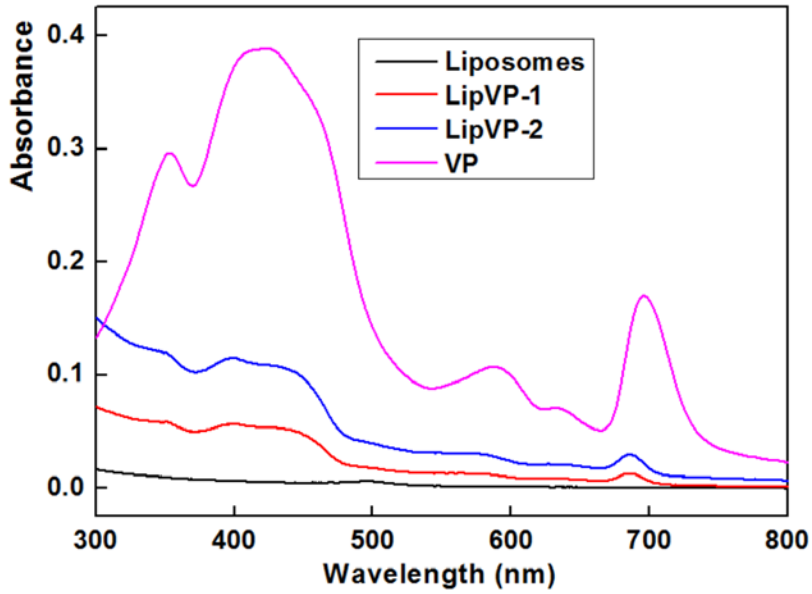

Figure S1 Absorbance spectra of liposomes alone, lipVP-1 (5.55 μg/mL), lipVP-2 (55.5 μg/mL) and free VP molecules.

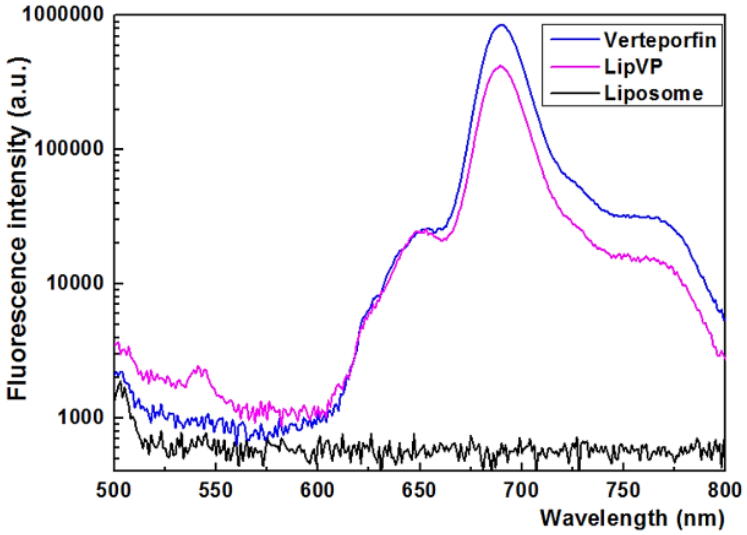

Figure S2 Fluorescence spectra of liposomes alone, lipVP and free VP molecules under 425 nm excitation.

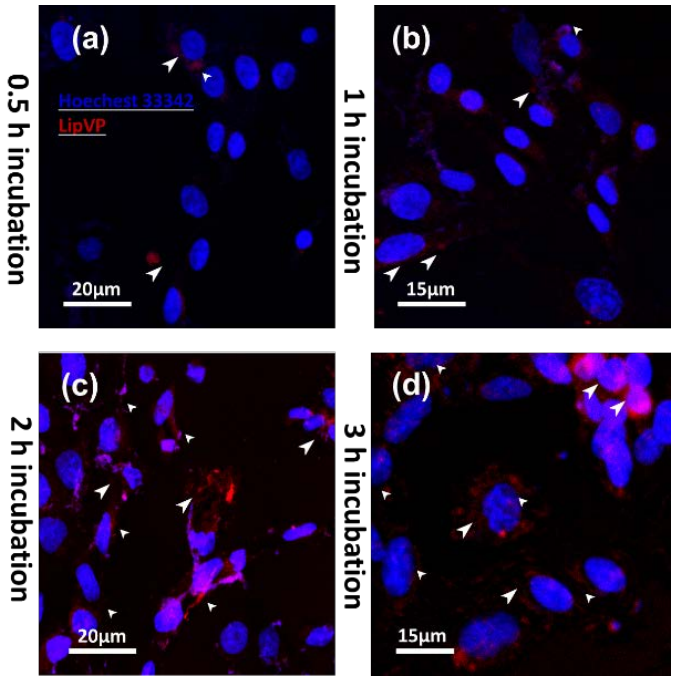

Figure S3 Cellular uptake. The confocal images of cellular uptake of lipVP in the serum-free medium at different incubation time points: (a) 0.5 h, (b) 1 h, (c) 2 h and (d) 3 h. White arrows refer to the lipVP nanoparticles surrounding the cell nucleus.

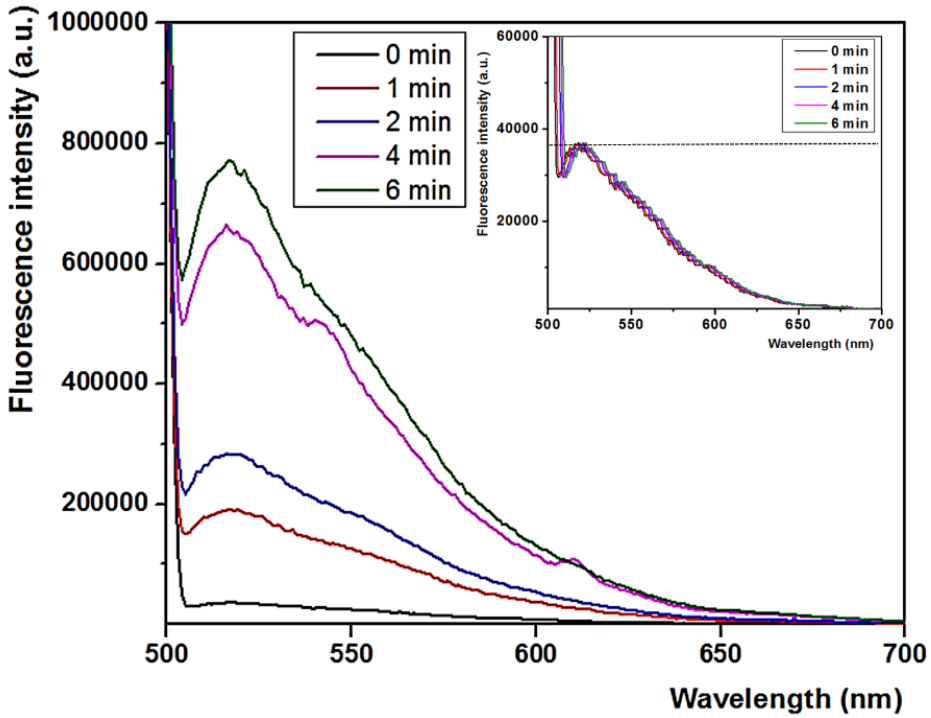

Figure S4 DNA release test. The release test of FAM labelled asODN encapsulated in lipVP and pure liposomes (inset) after UV illumination (365nm, 1.25mW/cm^2) for different time periods. The fluorescence intensity of FAM was measured at 425nm excitation.

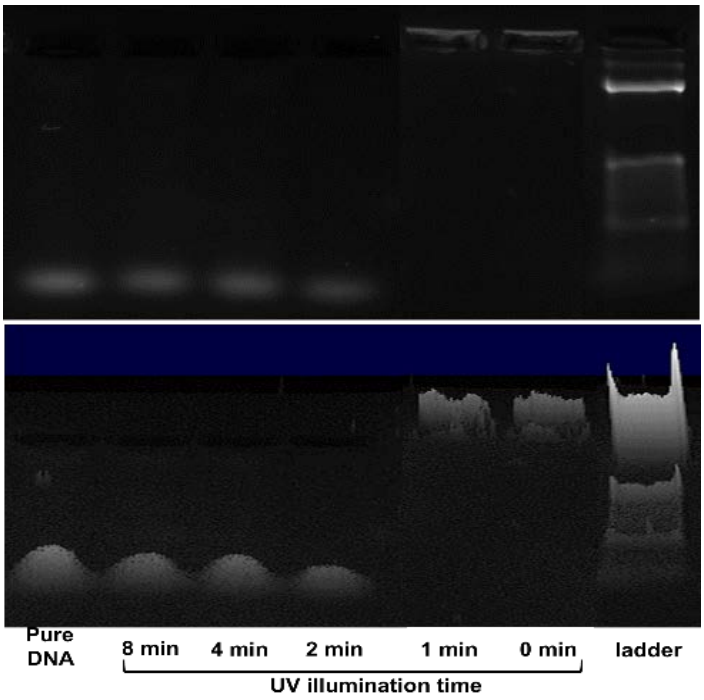

Figure S5 DNA damage assay. Agarose electrophoresis of lipVP/DNA complexes under different time of UV illumination. The below picture is the 3D version of the above.

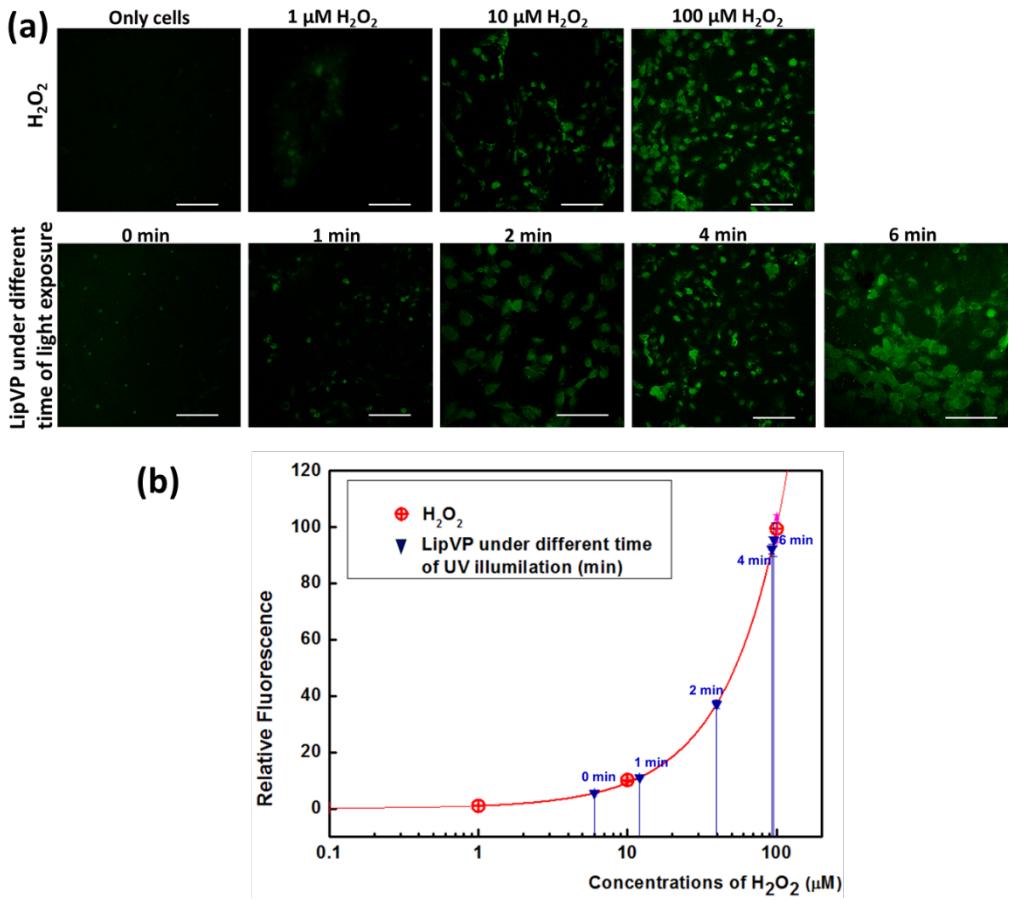

Figure S6 Detection of cellular ROS. (a) CLSM images of DCF fluorescence signal produced by the cellular ROS with and without light illumination. H_2O_2 solution with varying concentrations was added to cells in positive control groups. Scale bar, 120 μm. (b) Quantitative assessment of DCF fluorescence intensity after light treatment compared with the H_2O_2 positive controls. The amount of ROS was calculated from the fitting curve of H_2O_2-treated groups. LipVP generated different amount of ROS in transfected cells after 0, 1, 2, 4 and 6 min of illumination, which was equivalent to the amount produced with the introduction of 6.0 μM, 12.1 μM, 39.6 μM, 93.1 μM and 96.1 μM of H_2O_2, respectively.

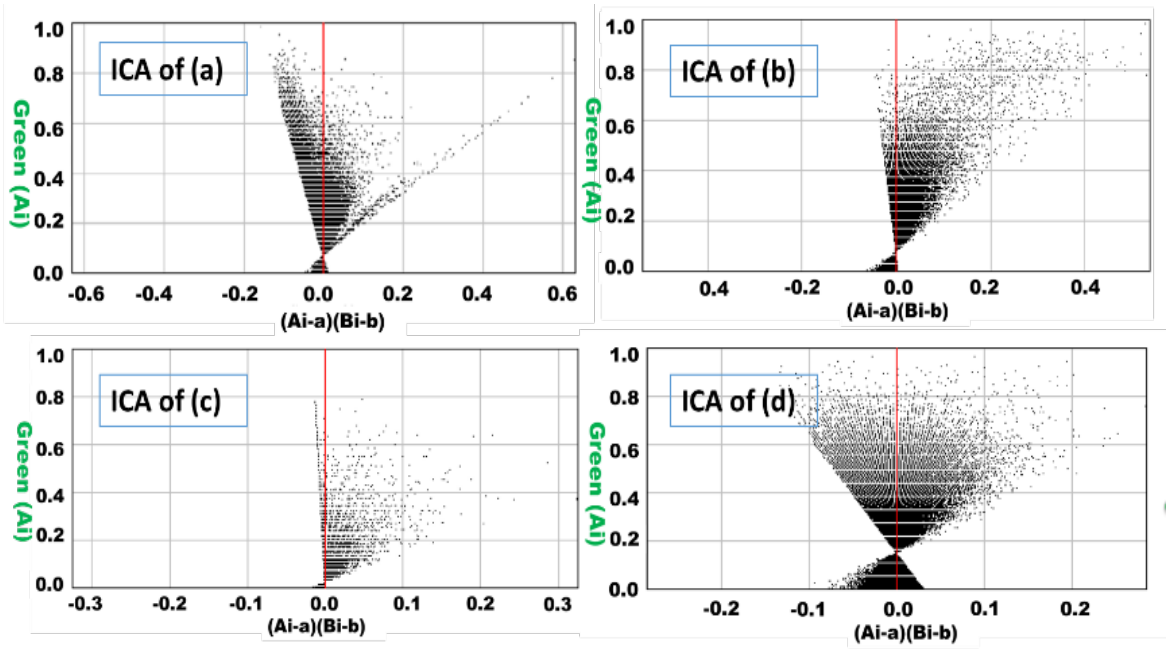

Figure S7 Intensity correlation analysis (ICA) of Figure4 a, b, c and d by using imageJ JACoP.

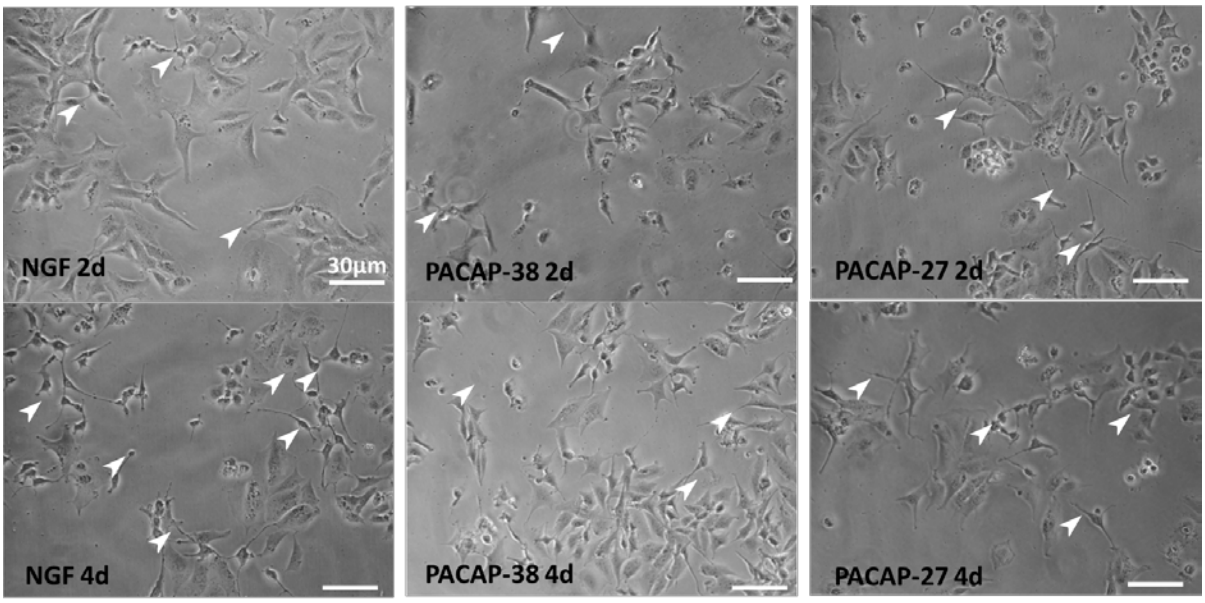

Figure S8 Phase contrast images of cell differentiation induced by NGF, PACAP-38 and PACAP-27 with 2-day and 4-day treatment. Scale bars: 30 μm. White arrows indicate selected typical neurites.

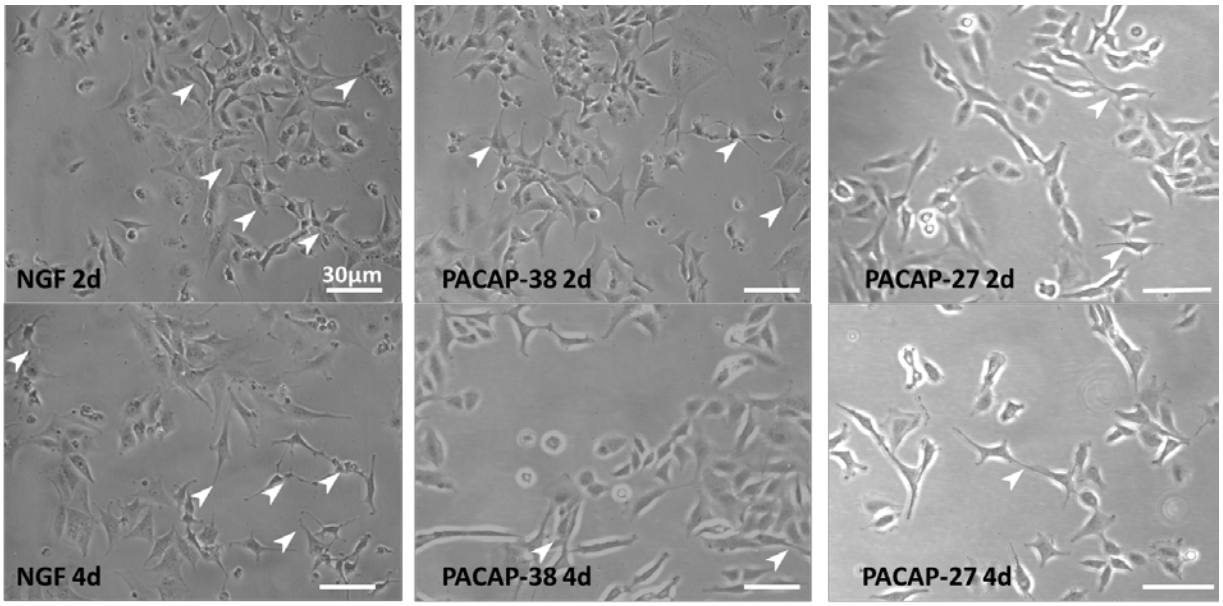

Figure S9 Phase contrast images of cell differentiation induced by NGF, PACAP-38 and PACAP-27 after transfection with lipVP-asODN complexes and UV illumination for 1 min. Scale bars: 30 μm. White arrows indicate selected typical neurites.

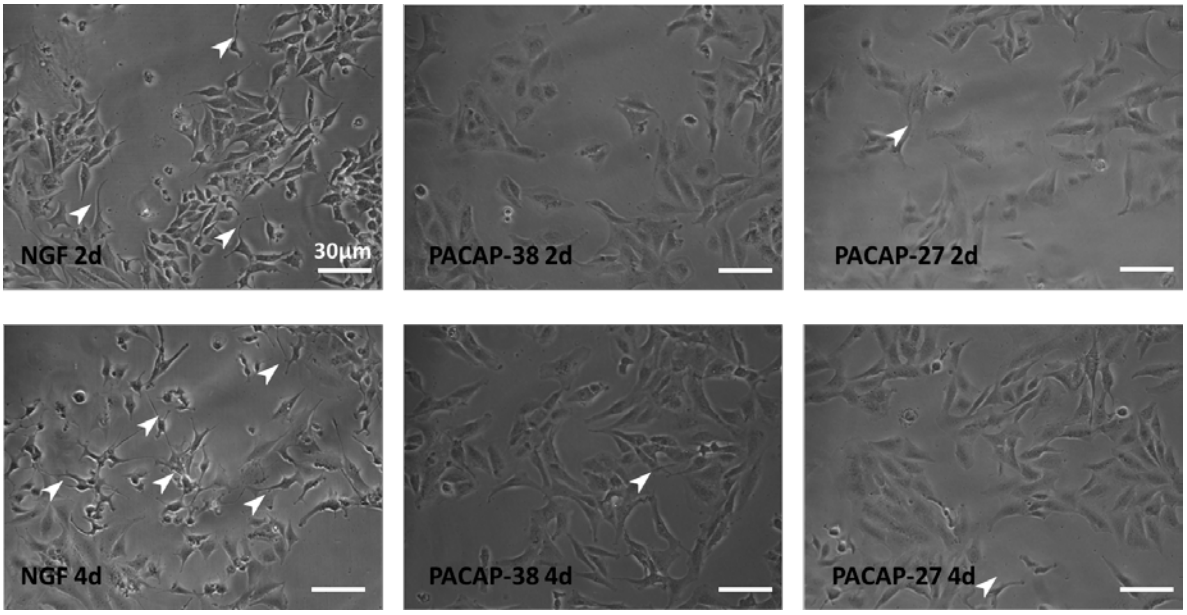

Figure S10 Phase contrast images of cell differentiation induced by NGF, PACAP-38 and PACAP-27 after transfection with lipVP-asODN complexes and UV illumination for 4 min. Scale bars: 30 μm. White arrows indicate selected typical neurites.

Supplemental methods

To determine if generated ROS can obviously damage genes or not, we evaluated the performance of DNA release in solution after light illumination using gel electrophoresis.In each vial, 0.5 μg of DNA was encapsulated into the as prepared lipVP to form the lipoplexes (lipVP/DNA at N/P ratio= 25), followed by 2, 4 and 8 min of UV illumination.