

Supplementary Information

A novel potent autophagy inhibitor ECDD-S27 targets vacuolar ATPase and inhibits cancer cell survival

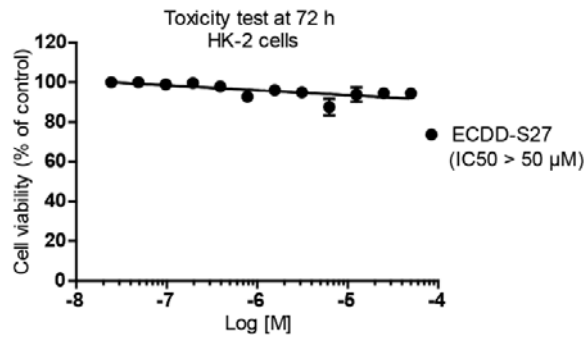
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Supplementary Methods

Cell toxicity assay

Human kidney normal cell HK-2 cells were obtained from ATCC. Cells were maintained in DMEM (Dulbecco's Modified Eagle Medium) supplemented with 10% FBS, 1% penicillin/streptomycin, and 1% HEPES at 37°C in 5% CO₂. At 24 h before treatment, HK-2 cells (1 x 10⁴ cells) were plated onto 96-well plates. Cells were then treated with DMSO (negative control) or different concentrations of ECDD-S27 for 72 h. Cell toxicity was then measured by using the MTT assay (Sigma-Aldrich) per manufacturer's recommendation. The absorbance at 570 nm was determined by Multi-Mode Microplate Reader (ENVISION, Perkin Elmer).

Supplementary Figures



Supplementary Fig. S1. ECDD-S27 is not toxic to human kidney normal HK-2 cells. HK-2 cells were treated with DMSO (negative control) or varied concentrations of ECDD-S27 for 72 h and cell toxicity was measured by the MTT assay. Data are mean \pm SEM done in triplicate; the results were expressed relative to the DMSO control, defined as 100%.

Images of immunoblotting

