Supplementary figure 1:

A: Chemical structure of TW-37 and TW-37a.

B: The levels of Bcl-2, Bcl-xL and Mcl-1 expression were compared between a panel of pancreatic cancer cell lines. The expression of protein was assayed by Western blot analysis.

C: Top, Dose responses of ApoG2 on growth of pancreatic cancer cells. Cells were seeded in 96-well plates at 5,000 cells per well and treated with varied concentrations of ApoG2 for 72 hours. After treatment, cell densities were determined by MTT assay. Each value represents the mean ± SD (n = 6) of three independent experiments. *P < 0.05, **P < 0.01, compared to the control.

Bottom, cells were cultured in RPMI containing 5% FBS and exposed to different concentration ApoG2 for 72 hours. Apoptosis was measured by Histone DNA ELISA. Values are reported as mean ± SD. *P < 0.05, compared to the control.

D: The expression of Notch-1 and its ligand Jagged-1 was detected by Western blotting in BxPC-3 and Colo-357 pancreatic cancer cells treated with ApoG2 for 72 hours.

Supplementary figure 2:

A: Effect of TW-37 on cell apoptotic death. Characterization of apoptosis was carried out after propidium iodide (PI) and Annexin V-FITC staining with apoptosis detection kit followed by flow cytometric analysis after 48 h of 500nM TW-37 treatment in BxPC-3 and Colo-357 cells. Briefly, 5 x 10^5 cells were treated with TW-37 for 48 h, and subjected to Annexin V staining. The cells were washed in PBS, resuspended in 100 µL of binding buffer containing a FITC-conjugated anti–Annexin V antibody, and then analyzed with a FACScalibur flow cytometer. The percentage of apoptotic cells increased from 5-6 % in the control to 12-14 % in both BxPC-3 and Colo-357 cell lines.

B: Notch-1 expression was up-regulated by Bcl-2 cDNA. Intracellular Notch-1 was increased in Bcl-2 cDNA transfected PC-3 and MCF-7 cells, compared to control transfected cells. Bcl-xL cDNA transfected PC-3 cells also showed high expression of Notch-1 activity and Hes-1.

Supplementary figure 3:

Schematic representation of the molecular effects of TW-37 on the Notch signaling network.