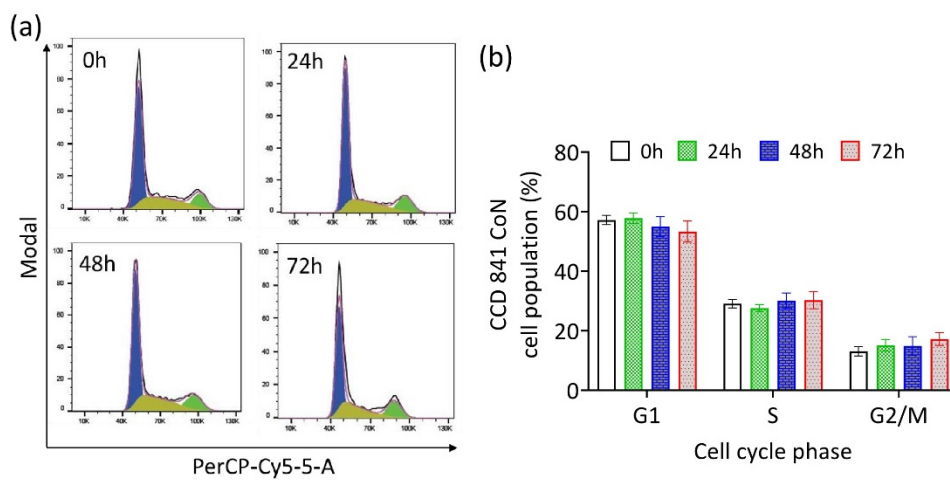
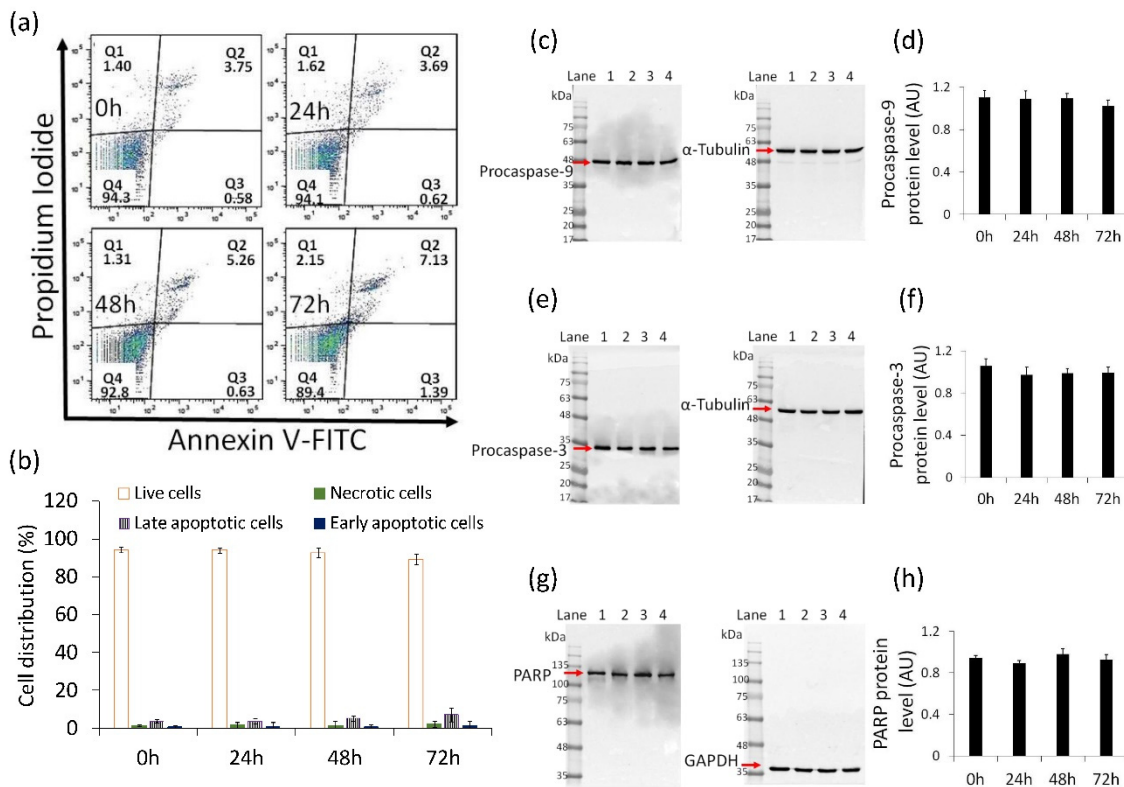


Colorectal Cancer Apoptosis Induced by Dietary δ -Valerobetaine Involves PINK1/Parkin Dependent-Mitophagy and SIRT3

SUPPLEMENTARY FIGURE



Supplementary Figure S1. Effects of δ VB on normal colon cell cycle. (a) Representative cell cycle distribution and (b) analysis of CCD 841 CoN normal colon cell line. CCD 841 CoN cells were treated with 1.5 mM δ VB for 24, 48 and 72 h. Cell distribution was assessed by flow cytometry collecting PI fluorescence as FL3-A (linear scale). For each sample at least 10,000 events were registered and analysis performed by using ModFIT software.



Supplementary Figure S2. Effects of δ VB on normal colon cell death. (a) Representative dot plots and (b) analysis of annexin V-FITC and PI-stained CCD 841 CoN normal colon cell line. CCD 841 CoN cells were treated with 1.5 mM δ VB for 24, 48 and 72 h. Cell viability/death was assessed by flow cytometry where at least 10,000 events were acquired. Q1: necrotic cells; Q2: late apoptotic cells; Q3: early apoptotic cells; Q4: viable cells. Data are expressed as mean \pm SD of n = 3 experiments. Representative full-length blots of Western blotting analysis of procaspase-9 (c,d), procaspase-3 (e,f) and PARP (g,h) in CCD 841 CoN cells. Lane 1 = 0 h; lane 2 = 24 h, lane 3 = 48 h, lane 4 = 72 h. Before lane 1; molecular weight markers (G266, Applied Biological Materials Inc., Richmond, BC, Canada). Protein expression was calculated, after normalization with internal control (α -tubulin or GAPDH), with Image J software and results expressed as arbitrary units (AU).