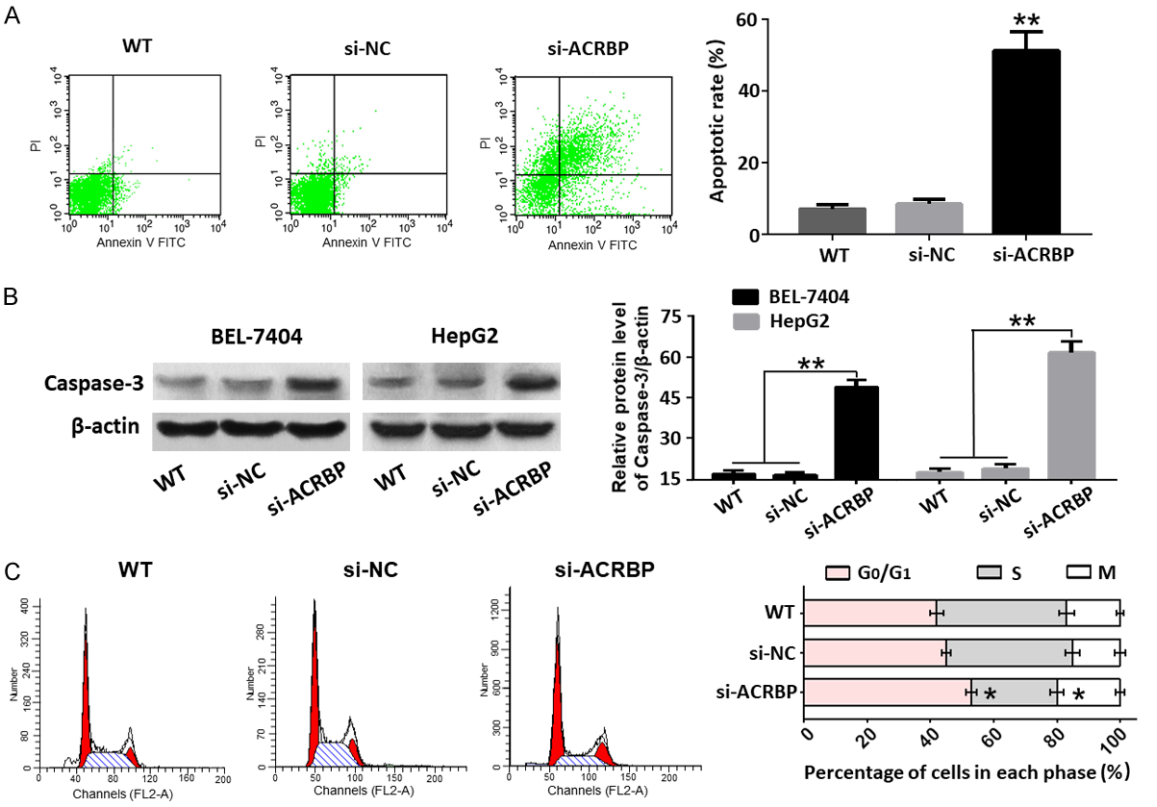
Table S1. Primers used in this assay

Gene	Description	Sequence (5'-3')	Amplificant (bp)
Human ACRBP	RT-PCR (F)	AAGGACAGGGGACTAAGGAG	604
Human ACRBP	RT-PCR (R)	CCGTACAAATCCAGCCCGTA	
Mouse ACRBP	RT-PCR (F)	ATGATGAATCTAGCTGCTGG	951
Mouse ACRBP	RT-PCR (R)	TCACAATTTCCTGTACCTGC	
Mouse β-actin	RT-PCR (F)	ATGGAGAAGATCTGGCACCA	709
Mouse β-actin	RT-PCR (R)	TAATCTCCTTCTGCATCCTGTC	
Mouse ACRBP (5 CpGs)	Pyrosequencing (F)	GTAGTTAGATTTATTATGGTTTGGGAAGAT	171
Mouse ACRBP (5 CpGs)	Pyrosequencing (R)	ATAACTCCTAACATAACCCCCTTATCC	
Mouse ACRBP (3 CpGs)	Pyrosequencing (F)	GGGGTTATAGTTAGGAGAAATTAGG	171
Mouse ACRBP (3 CpGs)	Pyrosequencing (R)	AACCAAAAACAAAAAAACCATATCAA	
Human ACRBP (4 CpGs)	Pyrosequencing (F)	GAGGTTGAGGTAGGAGAATTATT	150
Human ACRBP (4 CpGs)	Pyrosequencing (R)	ATAACAAACAACTACCTTCACTTAAC	

Table S2. Antibodies used in flow cytometry

Target	Label	Manufacturer (Catalog number)	Manufacturer (Catalog No) of isotype control
CD11c	PE, FITC, PERCP-CY5	BD Pharmingen (553802, 553801, 560584)	BD Pharmingen (553954, 553953, 560554)
CD40	FITC	BD Pharmingen (553790)	BD Pharmingen (553929)
CD86	FITC	BD Pharmingen (561962)	BD Pharmingen (553929)
CD80	PERCP-CY5	BD Pharmingen (560526)	BD Pharmingen (560562)
CCR7	PERCP-CY5	BD Pharmingen (560812)	BD Pharmingen (550765)
MHC I	FITC	Abcam (ab25030)	Abcam (ab18455)
MHC II	PE	Abcam (ab25585)	BD Pharmingen (556653)
CD8α	PE	BD Pharmingen (553032)	BD Pharmingen (556653)
CD107α	PE	BD Pharmingen (558661)	BD Pharmingen (556653)



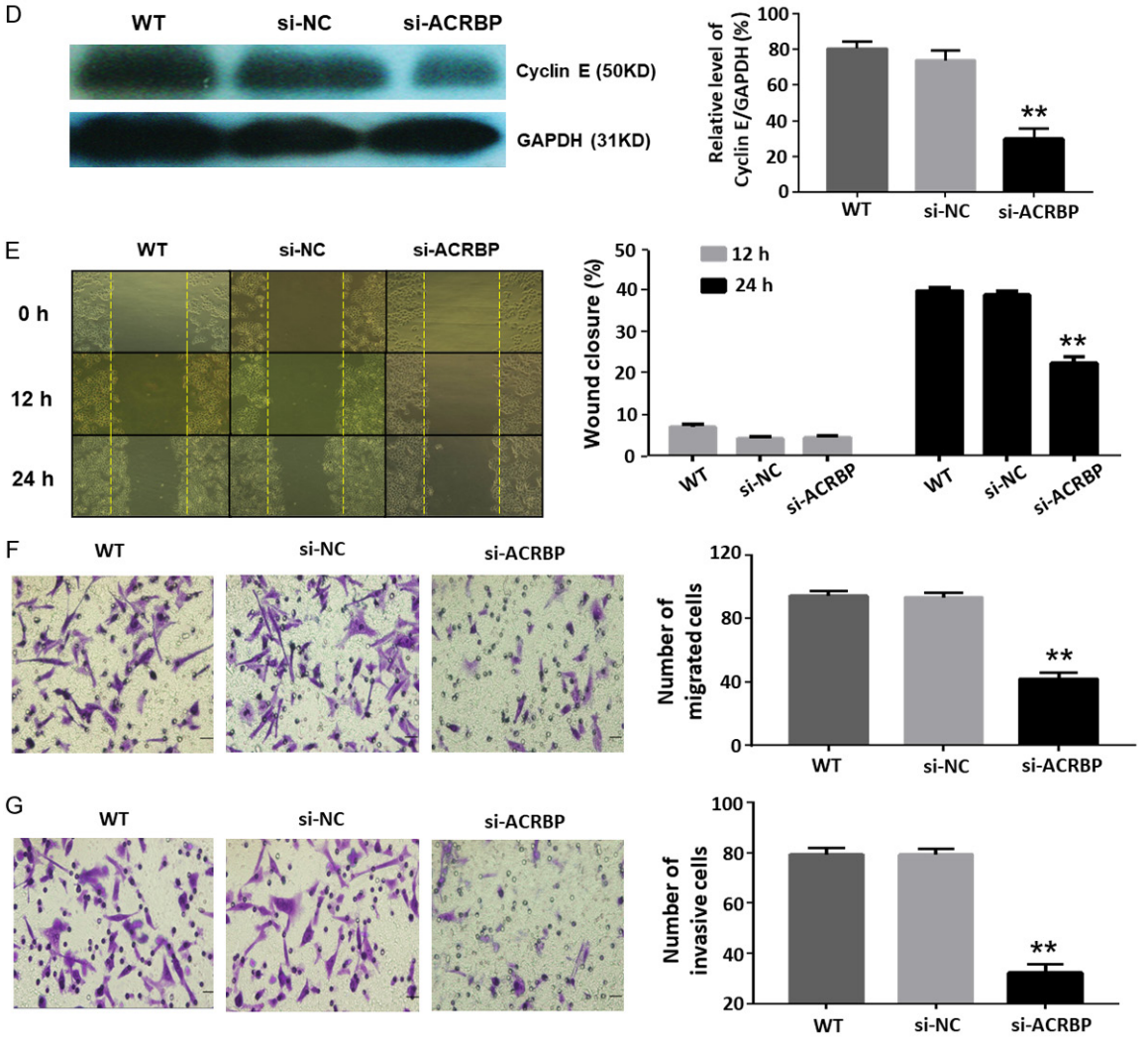


Figure S1. Downregulation of ACRBP by siRNA attenuated the malignant behaviors of BEL-7404 cells. (A) Apoptosis rates detected by flow cytometry were higher in cells transfected with ACRBP-specific siRNA as compared with control groups. (B) Higher Caspase-3 expression level was determined by immunoblotting in BEL-7404 and HepG2 cells transfected with ACRBP-specific siRNA as compared with control groups. (C) Cells transfected with ACRBP-specific siRNA showed G0/G1 phase arrest. (D) Immunoblotting was used to study the expression of Cyclin E in the cells. (E and F) Cells with lower ACRBP expression displayed lower capacity of migration demonstrated by wound healing assay (E) and transwell assay (F). (G) Invasion of cells with different treatments was determined by Matrigel transwell system. *, $P < 0.05$; **, $P < 0.01$.

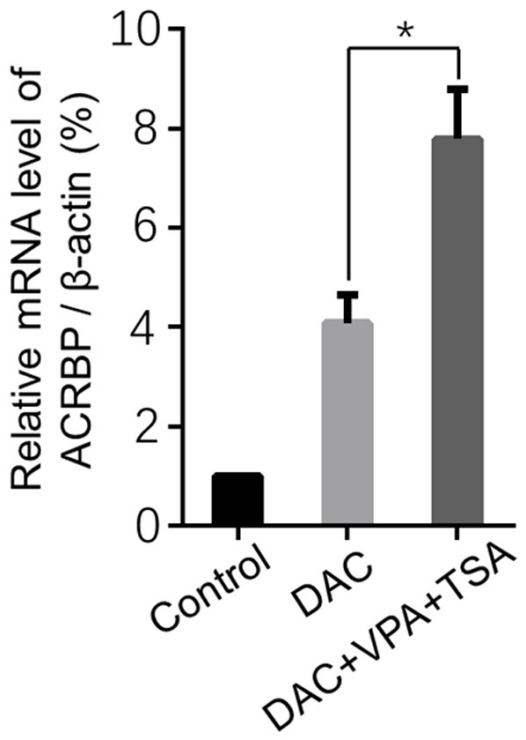


Figure S2. Relative mRNA expression of ACRBP in HepG2 cells treated with different drugs. DAC, target cells were treated with 5 μM DAC; DAC+VPA+TSA, target cells were treated with 5 μM DAC, 1 mM VPA and 2 μM TSA. *, $P < 0.05$.

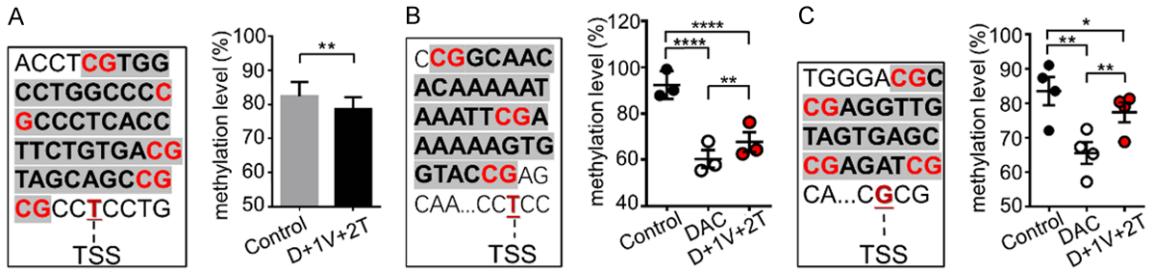
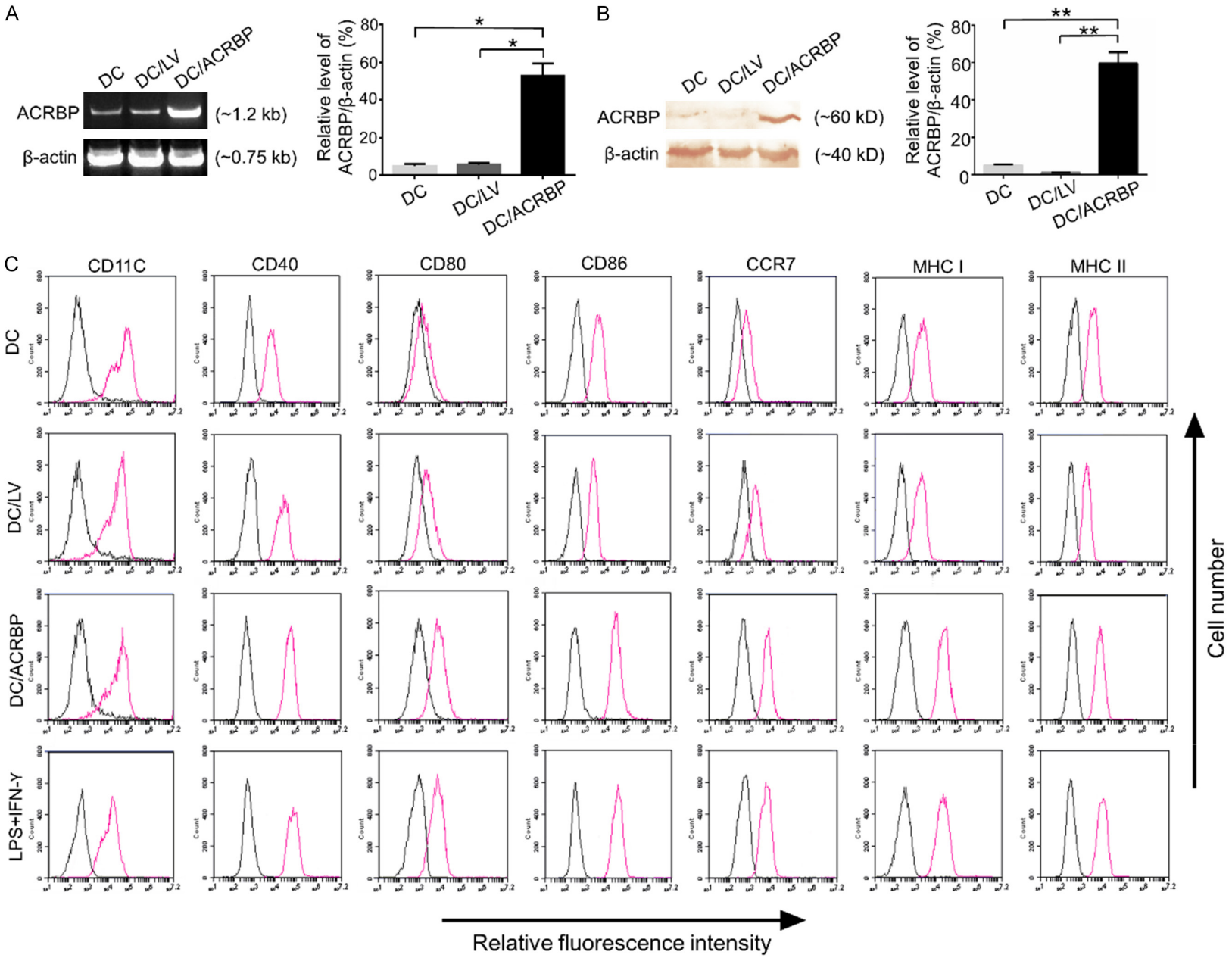


Figure S3. Methylation of ACRBP promoter was analyzed by pyrosequencing. (A) The average methylation level of 5 CpG sites in the ACRBP promoter of Hepa1-6 cells. Left panel, a 44-bp DNA fragment highlighted in grey (containing 5 CpG sites marked in red) was studied. Right panel, the 5 CpGs were demethylated after treatment with the triple drugs. (B) Average methylation levels of 3 CpG sites in the ACRBP promoter of Hepa1-6 cells. Left panel, a 37-bp DNA segment highlighted in grey (containing 3 CpG sites marked in red; 163 bp upstream of TSS) was examined. Right panel, average methylation levels of the 3 CpG sites were determined after Hepa1-6 cells were treated with DAC and triple-drug combination, respectively. (C) Average methylation levels of 4 CpG sites in the ACRBP promoter of HepG2 cells. Left panel, a 27-bp DNA fragment shaded in grey (containing 4 CpG sites colored in red; 188 bp upstream of TSS) was studied. Right panel, average methylation rates of the 4 CpG sites were evaluated after the HepG2 cells were cocultured with DAC and triple combined drugs, respectively. Each dot of the graphics in (B and C) (right panels respectively) represents the average of 3 independent biological replicates. *, $P < 0.05$; **, $P < 0.01$; ****, $P < 0.0001$.


A
DC
DC/LV
DC/ACRBP
ACRBP
(~1.2 kb)
β-actin
(~0.75 kb)
Relative level of ACRBP/β-actin (%)
B
(~60 kD)
(~40 kD)
C
CD11C
CD40
CD80
CD86
CCR7
MHC I
MHC II
LPS+IFN-γ
Cell number
Relative fluorescence intensity

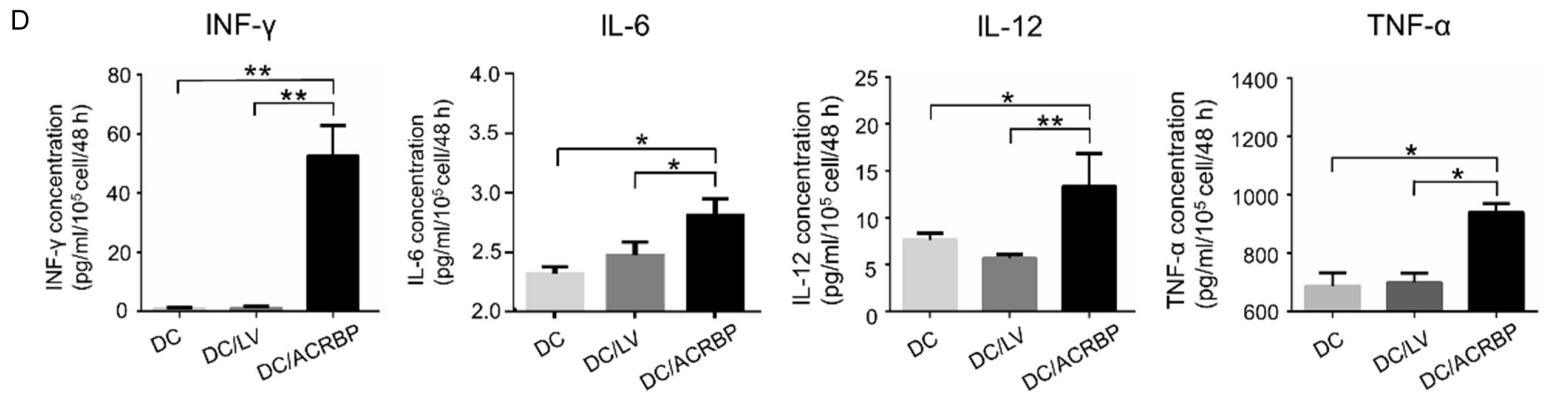


Figure S4. Overexpression of ACRBP promoted the maturation of DCs. (A and B) ACRBP expression of DCs transduced by LV-ACRBP (as described previously in "Materials and methods") was examined by RT-PCR (A) and immunoblotting (B). Left panel, representative images of ACRBP mRNA level (A) and protein level (B); right panels, relative ACRBP mRNA (A) and protein (B) expression normalized to β-actin. (C) Immunofluorescence analysis of surface markers expressed on DC2.4 cells under different treatments. Relative fluorescence activity of a specific monoclonal antibody is expressed in a red curve and that of isotype control is presented as a black one. (D) Secreted cytokines of DCs were studied by ELISA. *, $P < 0.05$; **, $P < 0.01$.