

**Supplementary Table 1 - Primers for Pyrosequencing**

	Forward	Reverse
1st/2nd CpG Island First Step	AGTTATGTATGTATGTGTTGTATATAGAGT	AAAAAATAATCCCCACTCTCCTATC
1st/2nd CpG Island Second Step	[Btn]TATATAGAGTAGATATATAGTTTATTAAG	CTATCTAATCCCTCCCTCTC
1st/2nd CpG Island Sequencing A	-----	CTTCCTCTCCAAAAATCTAA
1st/2nd CpG Island Sequencing B	-----	TTTCTTCTTCCCAATCTTTACC
1st/2nd CpG Island Sequencing C	-----	CCTCTCCTCCCCCAATTCCA
1st/2nd CpG Island Sequencing D	-----	CCAAATATCTTTTCTTCT
3rd CpG Island First Step	GAGGTGAGAGTGTTTTAGAT	CATAAACTTAATTTTCAATCTTAT
3rd CpG Island Second Step	GTTTATAATGATTAGTGTTTGTGGGAAAG	[Btn] AACCAAACCCCTCAAACAACAAAACCTA
3rd CpG Island Sequencing A	AGGTTAGAGAGGAAATTTTG	-----
3rd CpG Island Sequencing B	GGTTGAGAGTTAGGAT	-----

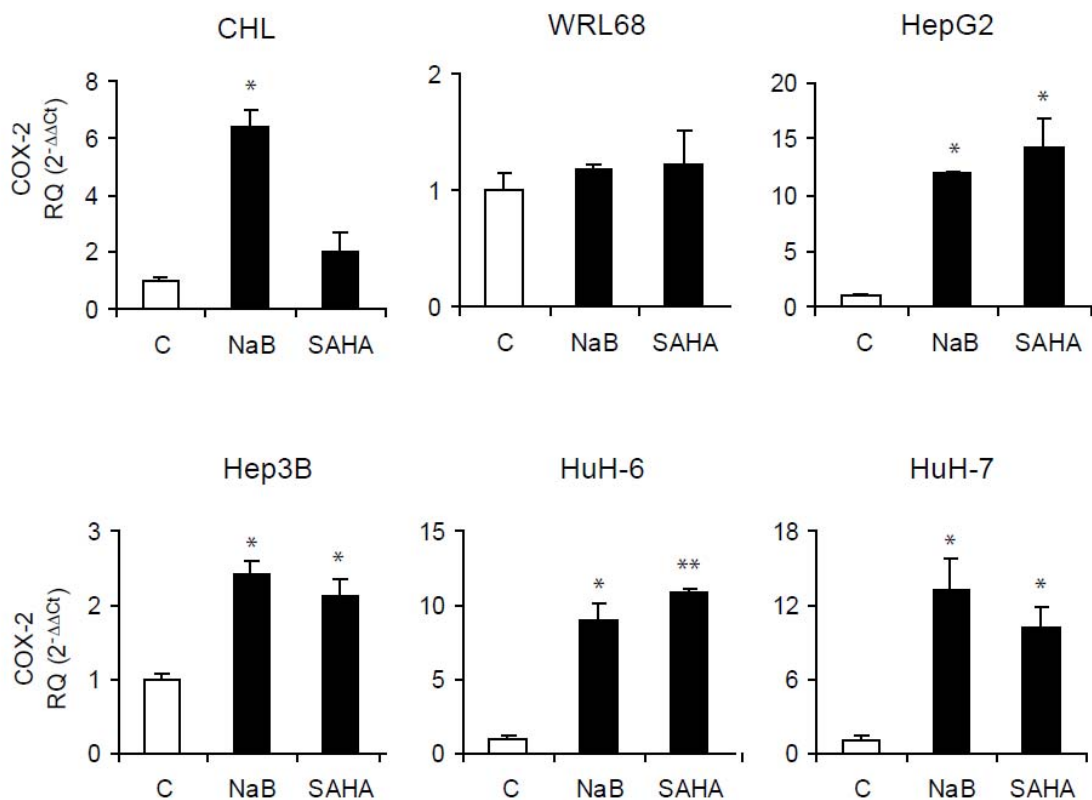
**Supplementary Table 2 - Primers for methylation-sensitive restriction assay (MSRA)**

	Forward	Reverse
CpG -431	CTTAACCTTACTCGCCCCAGTCT	AGAAGGACACTTGGCTTCCTC
CpG -372	GGAAGCCAAGTGTCTTCTGC	GGCAGGGTTTTTTACCCAC
CpG -138	AGCTTCCTGGGTTTCCGATT	CCCCACAAATTTTTCCCTC
CpG +75/84/98	GCCCTCAGACAGCAAAGCCTA	GGGAAAGCTGGAATATCCACG

**Supplementary Table 3 - Primers for Chromatin immunoprecipitation**

	Forward	Reverse
Distal Promoter	GGAAGCCAAGTGCCTTCTGC	TGATCGCCTTGGATGGGATA
Proximal Promoter	AGCTTCCTGGGTTTCCGATT	GGGCAGGGTTTTTTACCCAC
First Exon	CTCGGTTAGCGACCAATTGTC	CGTGCTCCTGACGCTCACT
First intron	AGCTTGGACCGCTAGAGTTTCG	GGGAAAGCTGGAATATCCACG

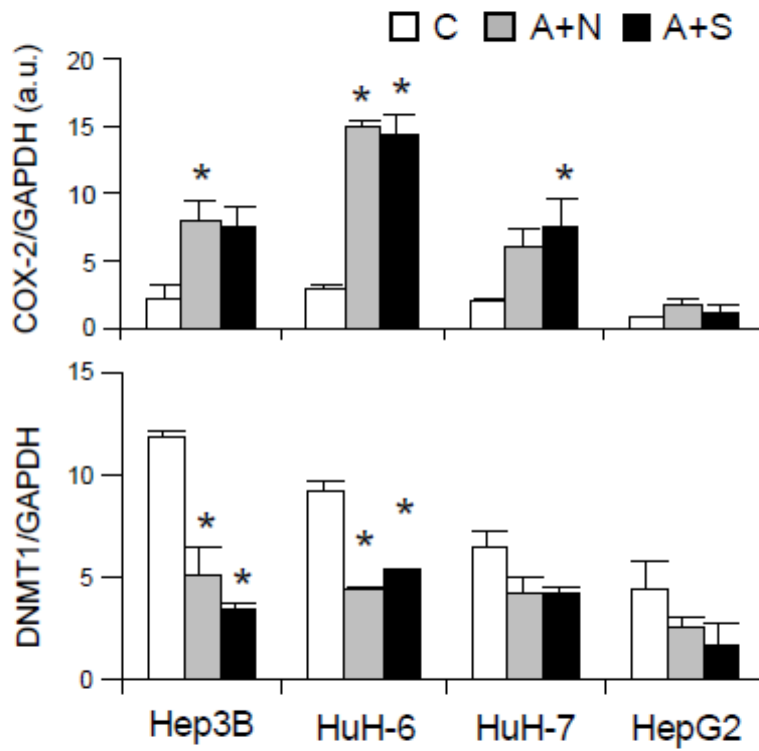
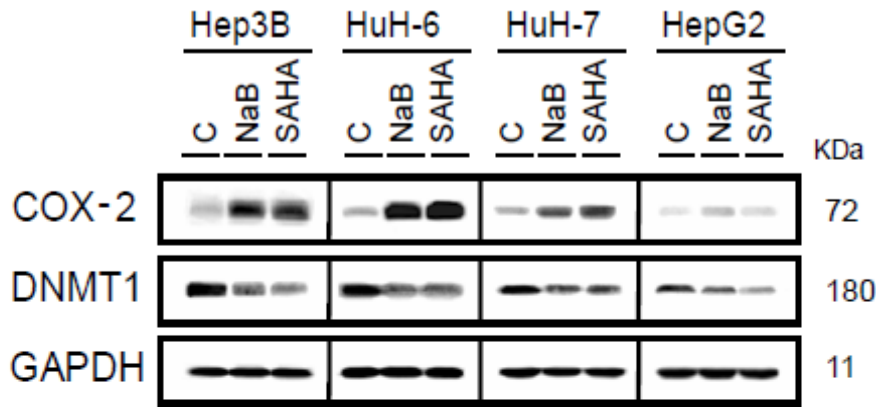
**Figure. S1- Expression of COX-2 increases after treatment of HCC cells with HDAC inhibitors.** Eight hours before the experiment HCC cells were change to 1% FBS and they were incubated without (white bars) or with 2.5 mM NaB or 2.5  $\mu$ M SAHA for 16 h. Total RNA was then isolated and mRNA levels for COX-2 and the internal control 18S were determined by qPCR. COX-2 mRNA amounts were calculated as RQ and normalized to the expression of ribosomal levels. Values represent fold change relative to each cell line without treatment. Data are reported as means  $\pm$  SD of three independent experiments. \*\* $p < 0.01$  and \* $p < 0.05$



**Figure. S2- Synergistic effect of HDAC inhibitors and AzadC on COX-2 expression.**

(A) HCC cells in 1% FBS were incubated without (white) or with 5  $\mu$ M AzadC and 2.5 mM NaB (A+N, grey) or AzadC and 2.5  $\mu$ M SAHA (A+S, black) for 48 h. Cellular extracts were prepared and protein was analyzed by Western blot. A representative Western blot analyzing COX-2 and DNMT1 protein levels is shown. The expression of target proteins was normalized to GAPDH. Each panel has an optimal exposition time. Densitometric analysis of COX-2 and DNMT1 protein levels is shown. Results are expressed as arbitrary units. (B) PGE<sub>2</sub> concentration was determined by enzyme immunoassay in the supernatant of the cells. Data are expressed as means  $\pm$  SD of three independent experiments. \* $p < 0.05$  vs. the corresponding cell line without treatment

A



B

