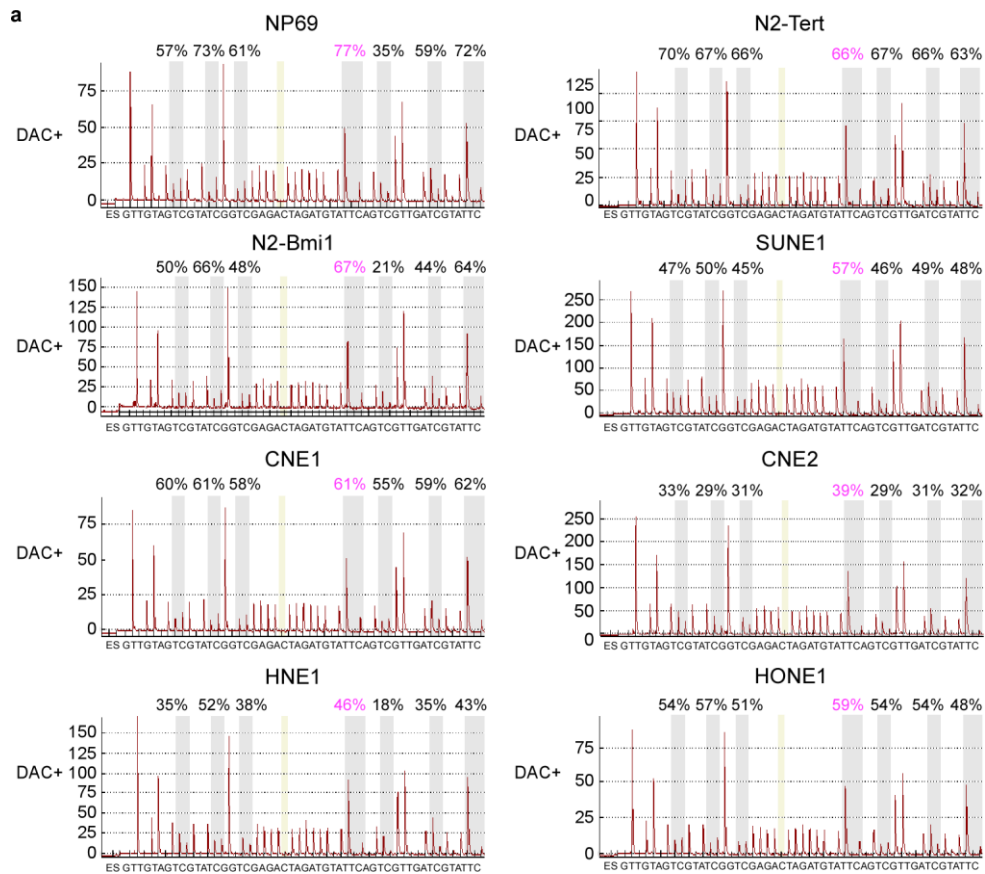
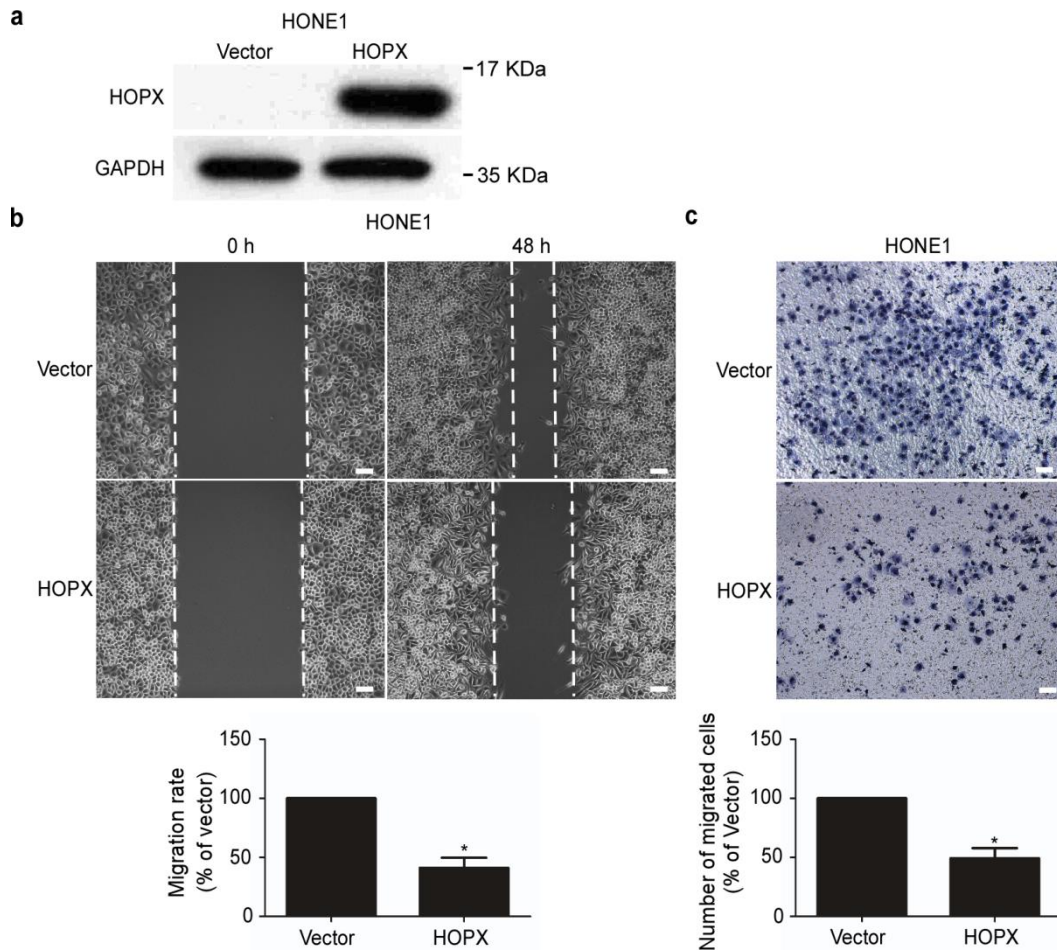


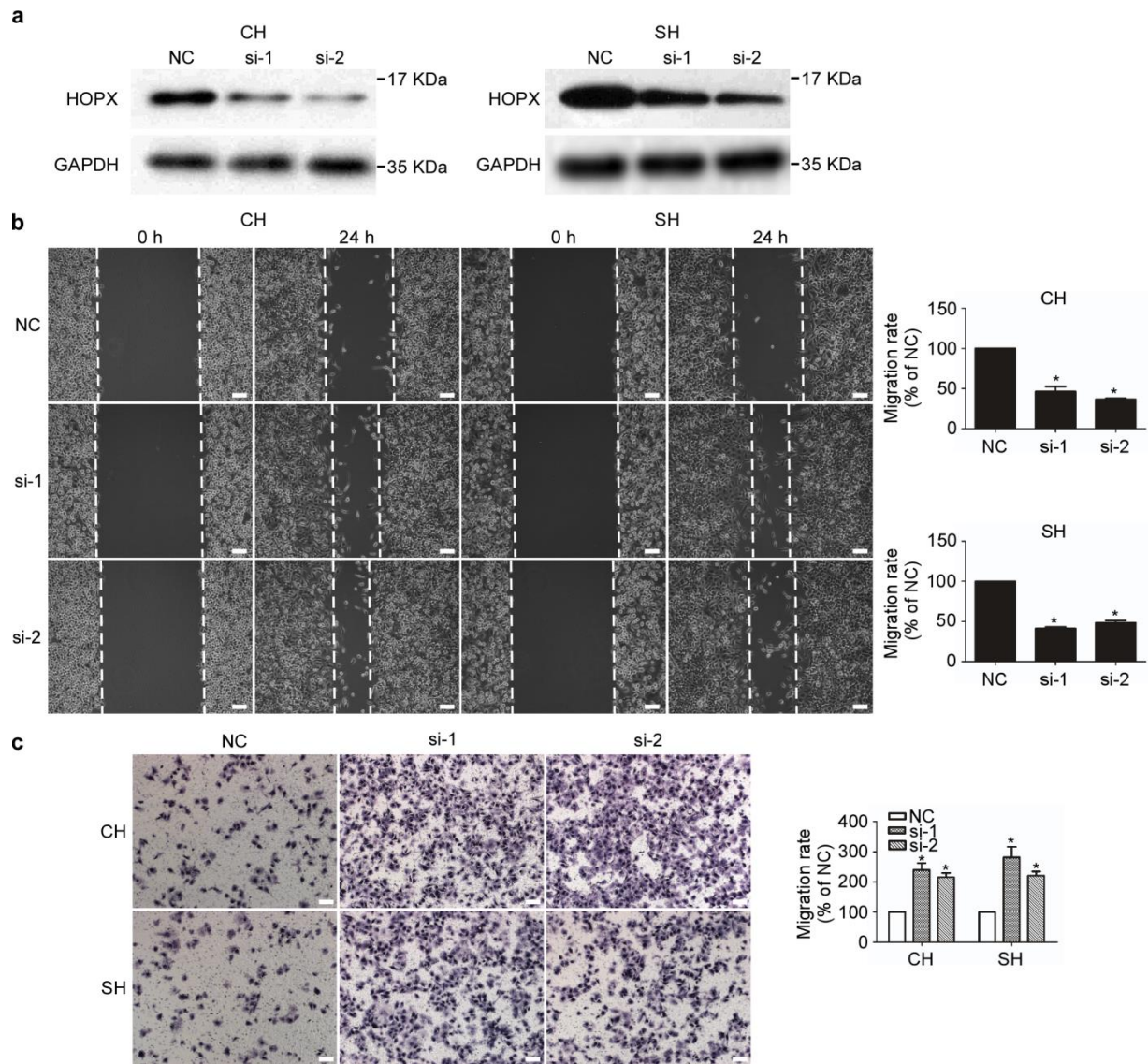
**Supplementary Figure 1. *HOPX* is hypermethylated in NPC.** (a) Methylation levels of *HOPX* in Normal (n = 24) and NPC (n = 24) tissues from the genome-wide methylation microarray data. Mean  $\pm$  s.d.; Student's *t*-tests. (b) Bisulfite pyrosequencing analysis of the *HOPX* promoter region in NPEC (NP69, N2-Tert and N2-Bmi1) and NPC (SUNE1, CNE1, CNE2, HNE1 and HONE1) cell lines. Magenta words: CG site of cg21899596. These data are representative of three independent experiments.



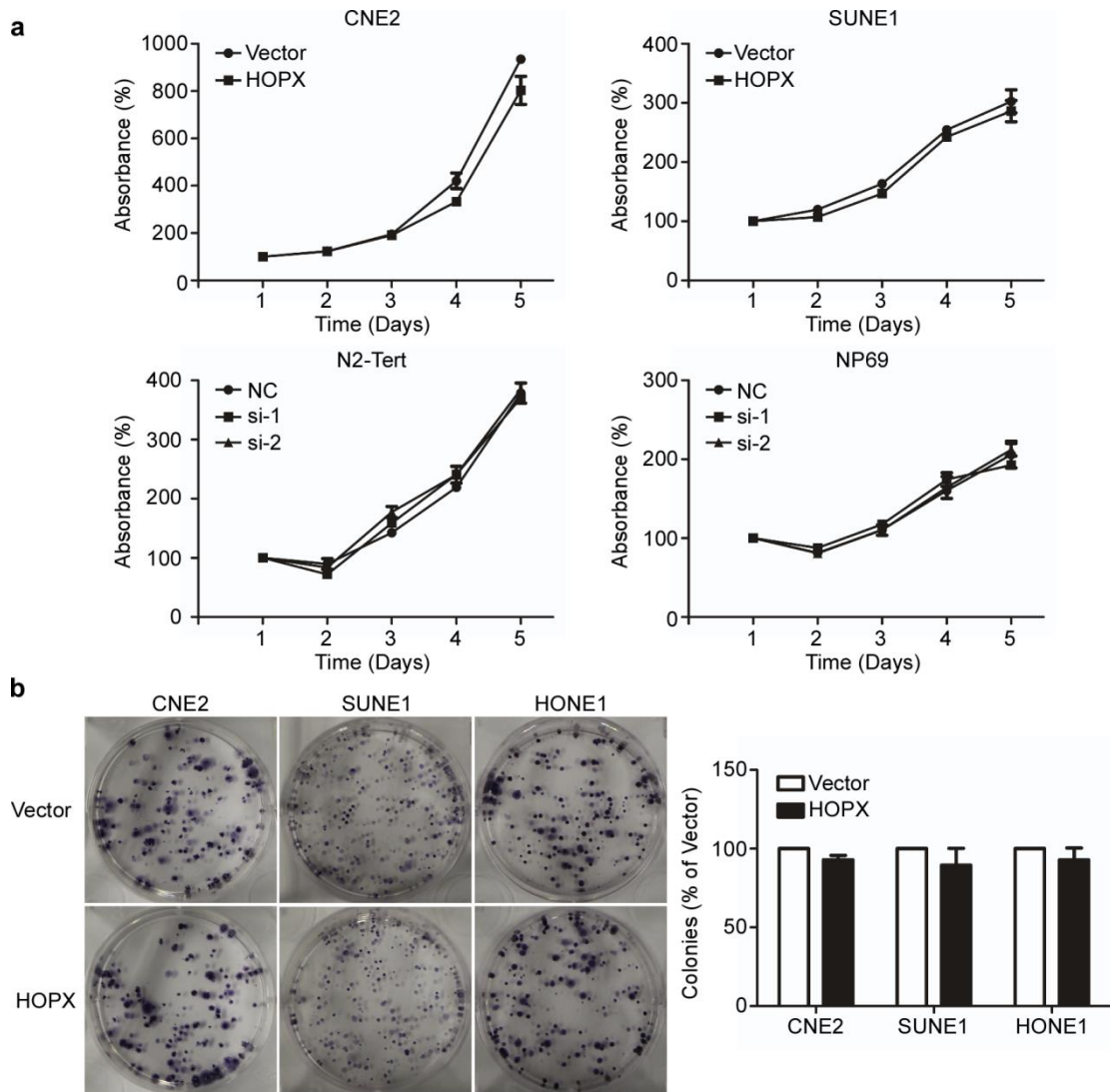
**Supplementary Figure 2. *HOPX* is hypermethylated in NPC cells.** (a) Bisulfite pyrosequencing analysis of the *HOPX* promoter region in NPEC (NP69, N2-Tert and N2-Bmi1) and NPC (SUNE1, CNE1, CNE2, HNE1 and HONE1) cell lines following treatment with DAC. Magenta words: CG site of cg21899596. These data are representative of three independent experiments.



**Supplementary Figure 3. HOPX suppresses HONE1 cell migration *in vitro*.** (a) The ectopic expression of HOPX in HONE1 cells was confirmed by western blotting. (b,c) Migration ability was measured using a wound healing assay (200 ×) (b) and Transwell assay without Matrigel (200 ×) (c) in HONE1 cells with the vector or HOPX overexpression. Scale bar: 100 μm; Mean ± s.d.; \*,  $P < 0.01$  compared with vector; Student's  $t$ -tests. These data are representative of three independent experiments.

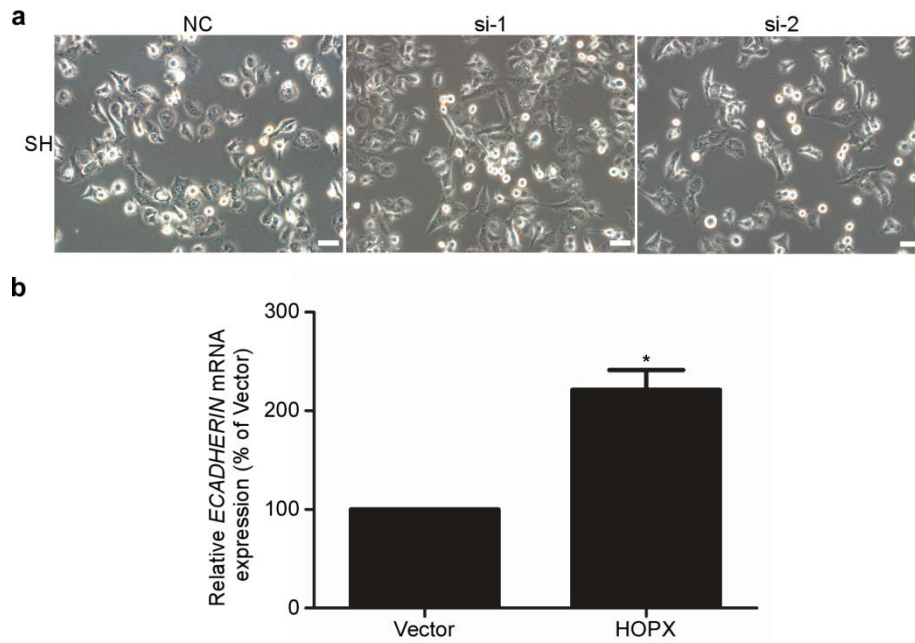


**Supplementary Figure 4. Silencing HOPX promotes NPC cell migration *in vitro*.** Control NC, or *HOPX*-siRNAs (si-1 and si-2) were used to knock down the expression of HOPX in CNE2 and SUNE1 cells with stable HOPX overexpression (CH and SH). **(a)** The silencing effects of HOPX in CH and SH cells were confirmed by western blotting. **(b,c)** Wound healing assay (200 ×) **(b)** and Transwell assay without Matrigel (200 ×) **(c)** were used to measure the migration abilities. Scale bar: 100 μm; mean ± s.d.; \*,  $P < 0.01$  compared with NC; Student's *t*-tests. These data are representative of three independent experiments.

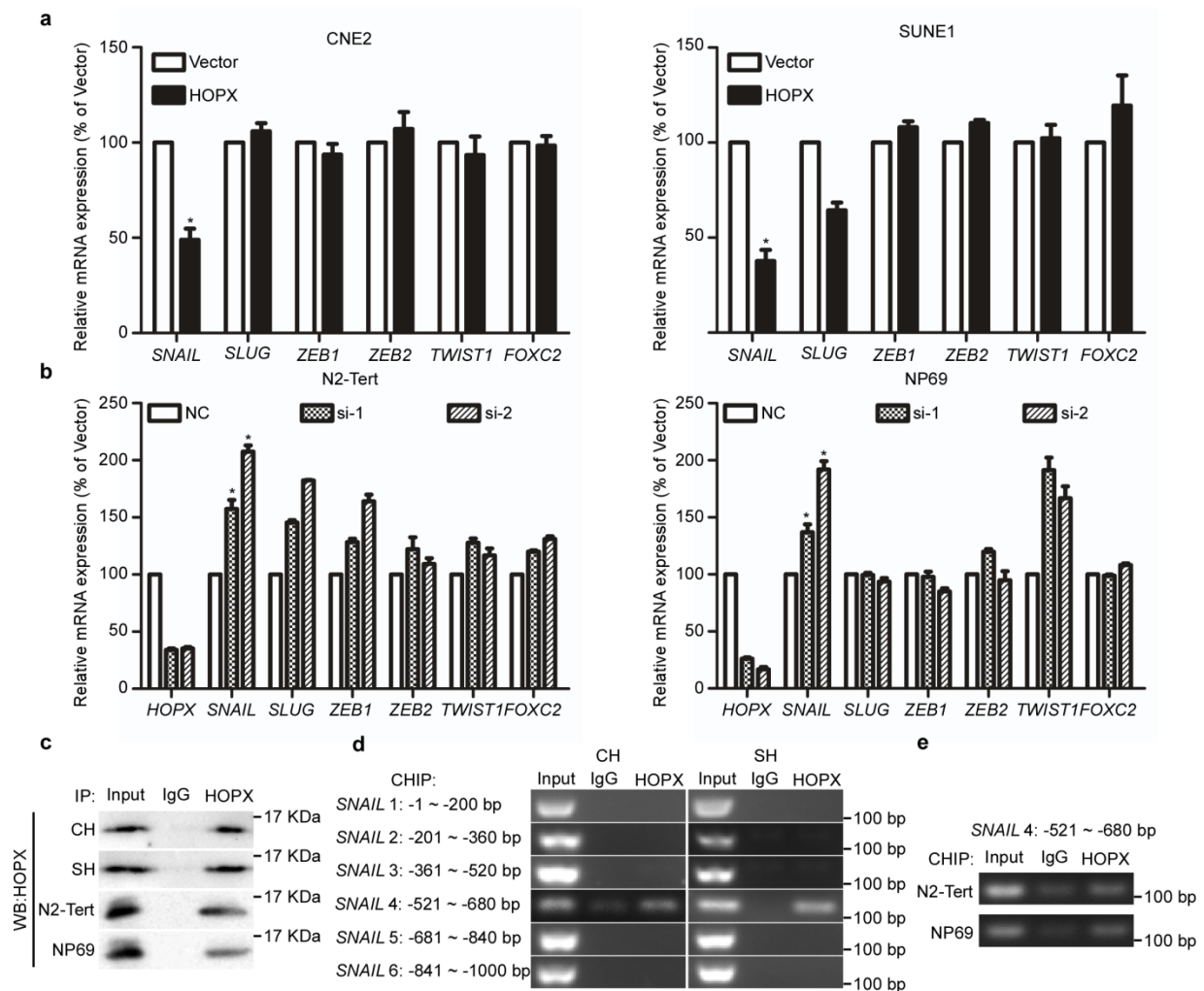


**Supplementary Figure 5. HOPX exhibits little effect on NPC cell proliferation *in vitro*.** (a) MTT assay was performed to examine the effect of HOPX on cell viability in CNE2 and SUNE1 cells stably overexpressed the vector or HOPX, or N2-Tert and NP69 cells transfected with the control NC or *HOPX*-siRNAs (si-1 and si-2). Mean  $\pm$  s.d.;  $P > 0.05$  compared with vector or NC; Student's *t*-tests. (b) Colony formation assay was used to examine the effect of HOPX on colonization in CNE2, SUNE1 and HONE1 cells with the vector or HOPX overexpression. Mean  $\pm$  s.d.;  $P > 0.05$  compared with vector; Student's *t*-tests. These data are representative of three independent experiments.

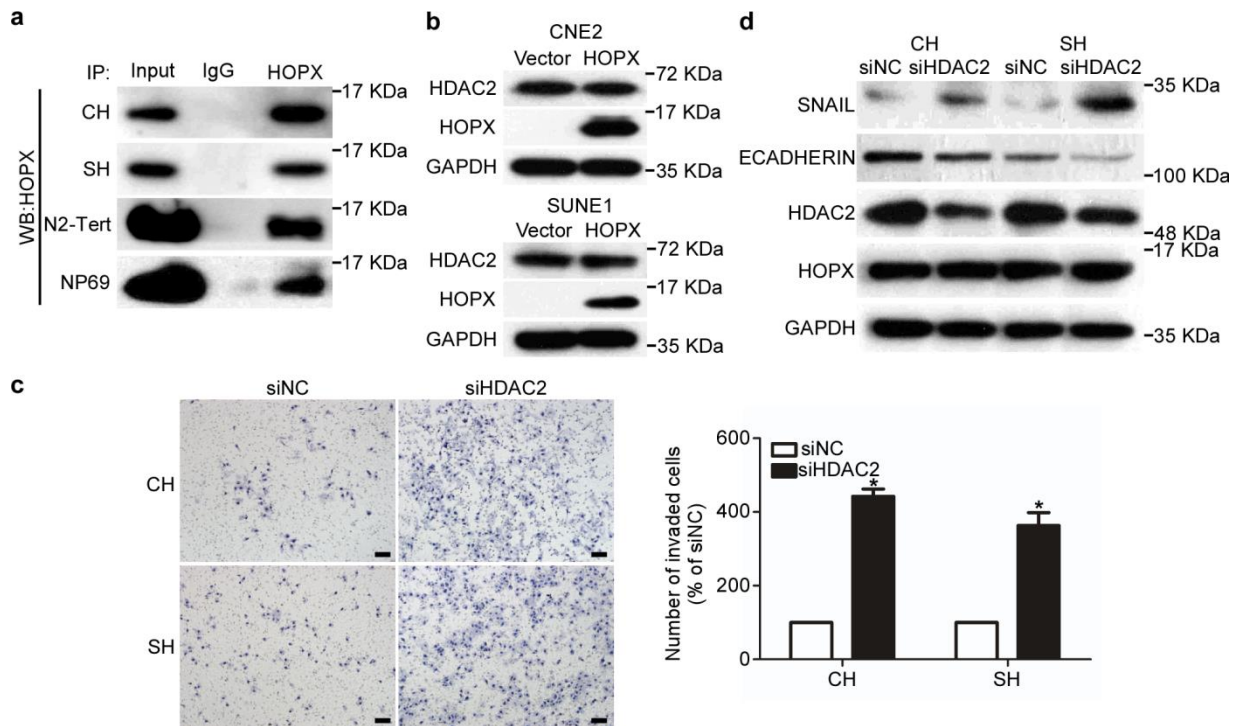




**Supplementary Figure 6. HOPX partially inhibits EMT in NPC cells.** (a) Phase contrast images (200 ×) of SH cells which were transfected with the control NC or *HOPX*-siRNAs (si-1 and si-2). SH cells indicated SUNE1 cells with *HOPX* overexpression. Scale bar: 100 μm. (b) The mRNA level of *ECADHERIN* was identified using a tumor metastasis PCR array in SUNE1 cells stably overexpressed the vector or *HOPX*. Mean ± s.d.; \*,  $P < 0.01$  compared with vector; Student's *t*-tests. These data are representative of three independent experiments.

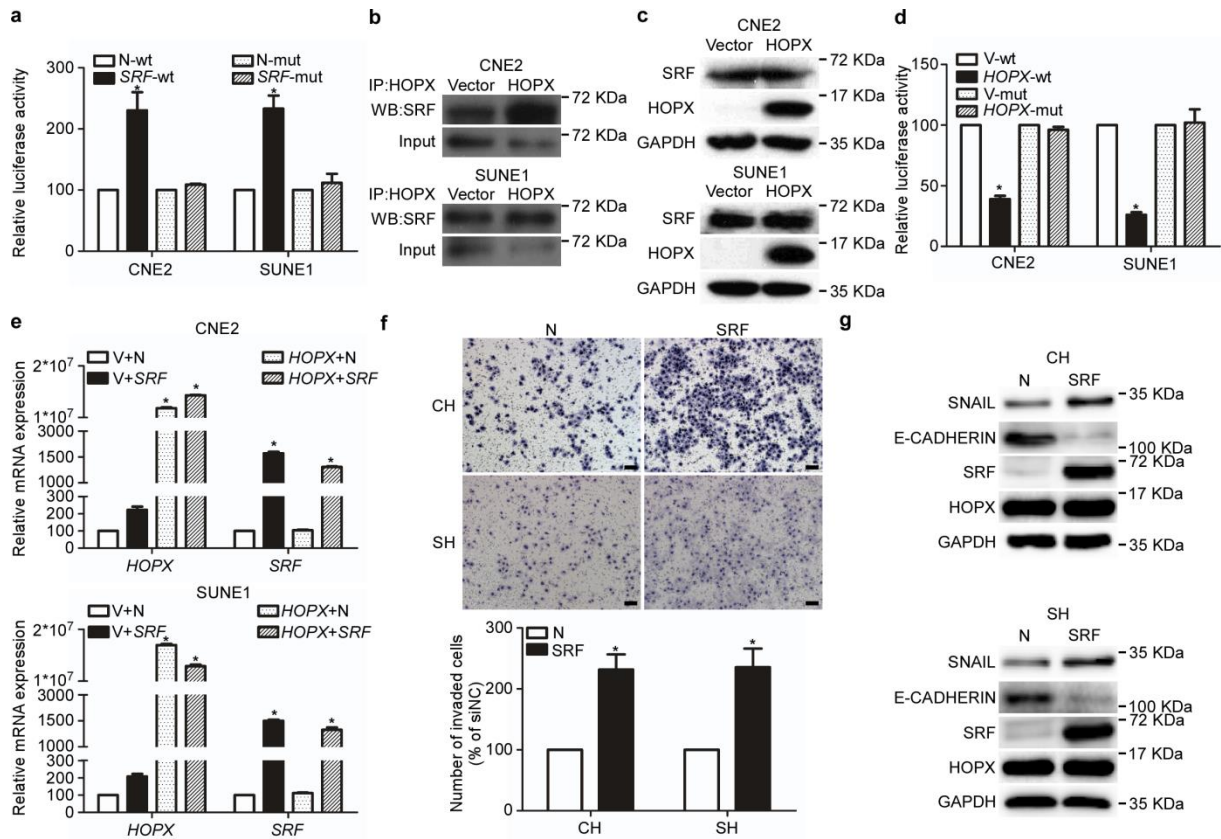


**Supplementary Figure 7. HOPX suppresses SNAIL expression in NPC.** (a,b) Real time RT-PCR was used to examine the mRNA level of EMT-TFs (*SNAIL*, *SLUG*, *ZEB1*, *ZEB2*, *TWIST1* and *FOXC2*) in CNE2 and SUNE1 cells stably overexpressed the vector or HOPX (a), or N2-Tert and NP69 cells transiently expressed control NC or HOPX-siRNAs (si-1 and si-2) (b). Mean  $\pm$  s.d.; \*,  $P < 0.01$  compared with vector or NC; Student's  $t$ -tests. (c) ChIP assay using an anti-HOPX antibody was performed to pull down HOPX. Western blotting (WB) was conducted to examine HOPX using an anti-HOPX Antibody. (d,e) ChIP-PCR assay was conducted to assess the enrichment of HOPX in the promoter regions of *SNAIL* in CH, SH, N2-Tert and NP69 cells. CH and SH cells indicated CNE2 and SUNE1 cells with HOPX exogenous expression. These data are representative of three independent experiments.

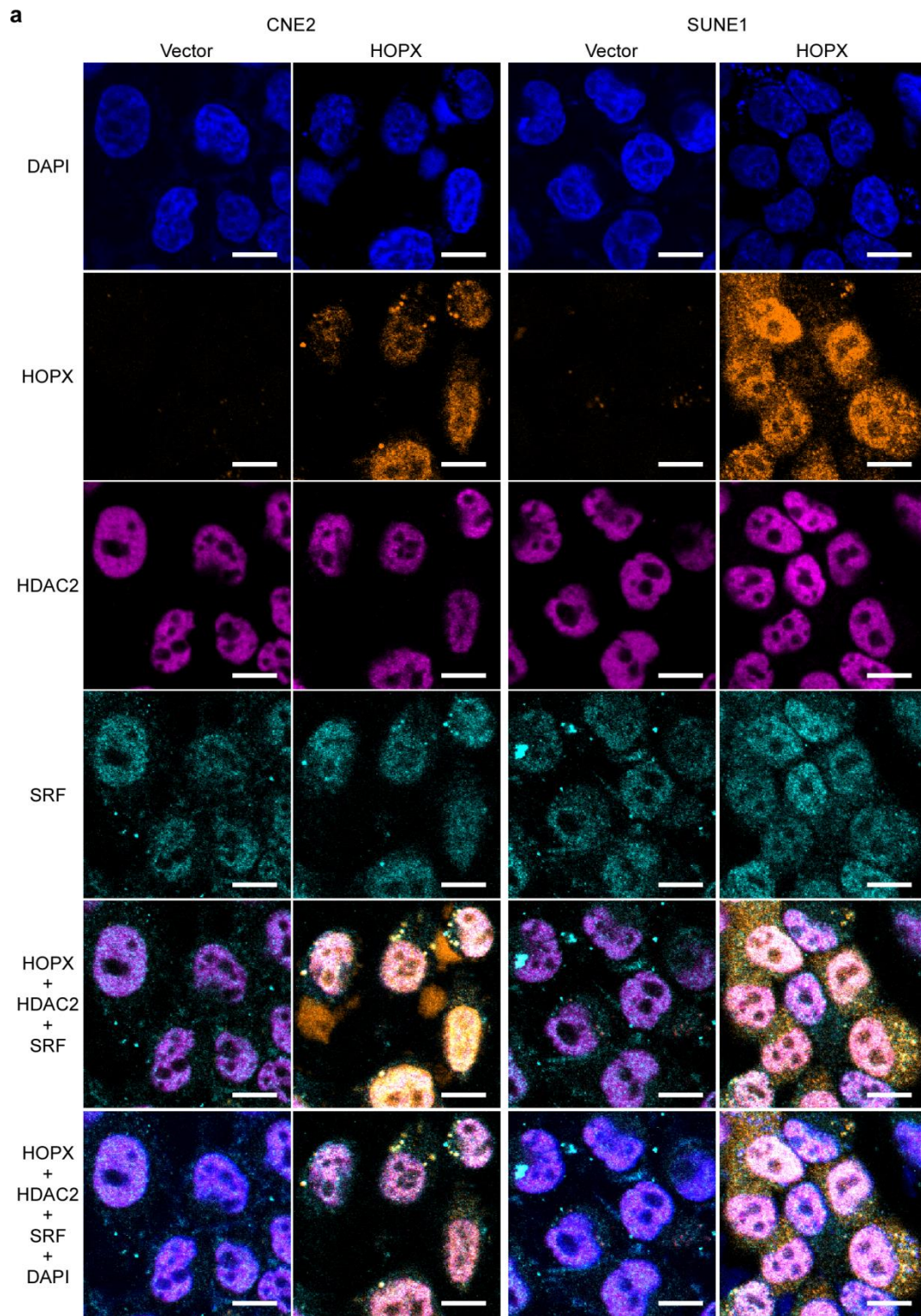


**Supplementary Figure 8. Silencing HDAC2 reverses the inhibitory effects of HOPX on invasiveness and EMT in NPC.** (a) IP assay using anti-HOPX antibody was performed to pull down HOPX. Western blotting (WB) was conducted to examine HOPX using an anti-HOPX Antibody. (b) Western blotting assay was used to assess the expression levels of HDAC2, HOPX and GAPDH. (c) Transwell assay with Matrigel (200  $\times$ ) was used to measure the invasive ability of CH and SH cells which were transfected with the control siNC or siHDAC2. Scale bar: 100  $\mu$ m; mean  $\pm$  s.d.; \*,  $P < 0.01$  compared with siNC; Student's  $t$ -tests. (d) Western blotting assay was used to examine the expression levels of SNAIL, ECADHERIN, HDAC2, HOPX and GAPDH in CH and SH cells which were transfected with the control siNC or siHDAC2. CH and SH cells indicated CNE2 and SUNE1 cells with HOPX exogenous expression. These data are representative of three independent experiments.

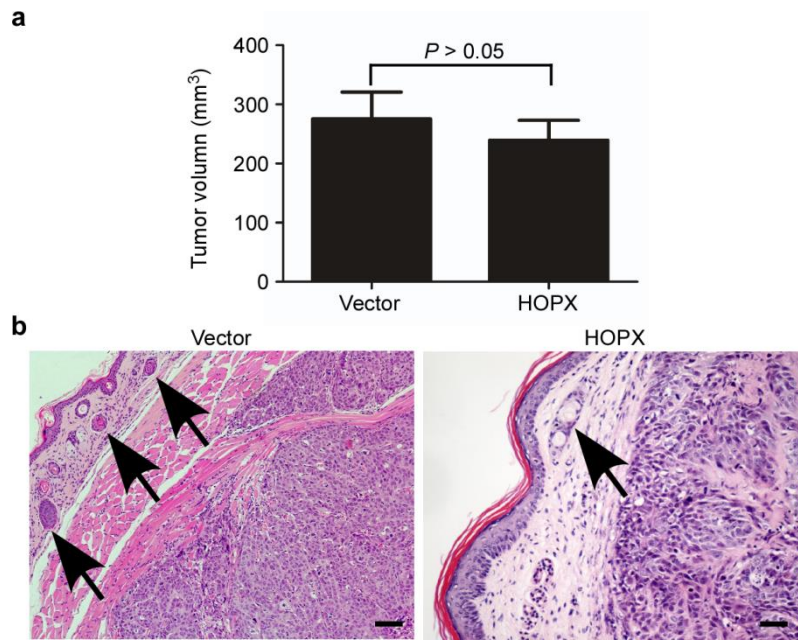




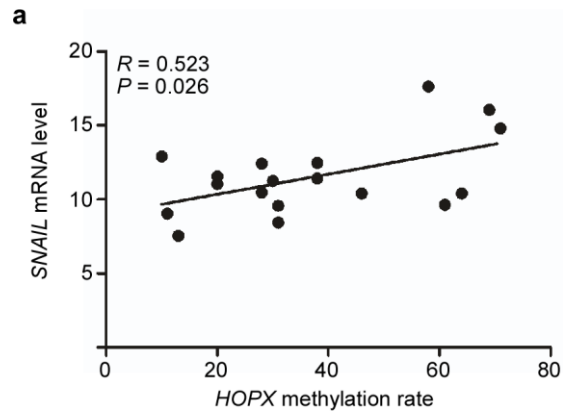
**Supplementary Figure 9. HOPX inhibits SRF-mediated *SNAIL* transcription in NPC.** (a) Wild type and mutant *SNAIL*-luciferase reporters were constructed and transfected with the control N or *SRF* construct in CNE2 and SUNE1 cells for 24 h. Luciferase reporter assay was used to detect the luciferase activity. Mean  $\pm$  s.d.; \*,  $P < 0.01$  compared with N-wt; Student's  $t$ -tests. (b) Co-IP assay was used to measure the interaction between HOPX and SRF in CNE2 and SUNE1 cells stably overexpressed the vector or HOPX. (c) Western blotting assay was used to assess the expression levels of SRF, HOPX and GAPDH. (d) Wild type and mutant *SNAIL*-luciferase reporters were constructed and transfected with the vector (V) or *HOPX* construct in CNE2 and SUNE1 cells for 24 h. Luciferase reporter assay was used to detect the luciferase activity. Mean  $\pm$  s.d.; \*,  $P < 0.01$  compared with V-wt; Student's  $t$ -tests. (e) CNE2 and SUNE1 cells were co-transfected with the *HOPX* and *SRF*. V and N were used as empty vectors of *HOPX* and *SRF*, respectively. Relative *SRF* and *HOPX* mRNA expressions were measured via real time RT-PCR. Mean  $\pm$  s.d.; \*,  $P < 0.01$  compared with V + N; Student's  $t$ -tests. (f) Transwell assay with Matrigel (200  $\times$ ) was used to measure the invasive ability of CH and SH cells which were transfected with the control N or *SRF* expression plasmids. Scale bar: 100  $\mu$ m; mean  $\pm$  s.d.; \*,  $P < 0.01$  compared with N; Student's  $t$ -tests. (g) Western blotting assay was used to examine the expression levels of *SNAIL*, *ECADHERIN*, *SRF*, *HOPX* and *GAPDH* in CH and SH cells which were transfected with the control N or *SRF* expression plasmids. CH and SH cells indicated CNE2 and SUNE1 cells with HOPX exogenous expression. These data are representative of three independent experiments.



**Supplementary Figure 10. HOPX is co-localized with HDAC2 and SRF in NPC cells. (a)** Immunofluorescence images (600 ×) of HOPX (orange), HDAC2 (magenta) and SRF (cyan) expression in CNE2 and SUNE1 cells stably overexpressed the vector or HOPX. Scale bar: 100 μm. These data are representative of three independent experiments.

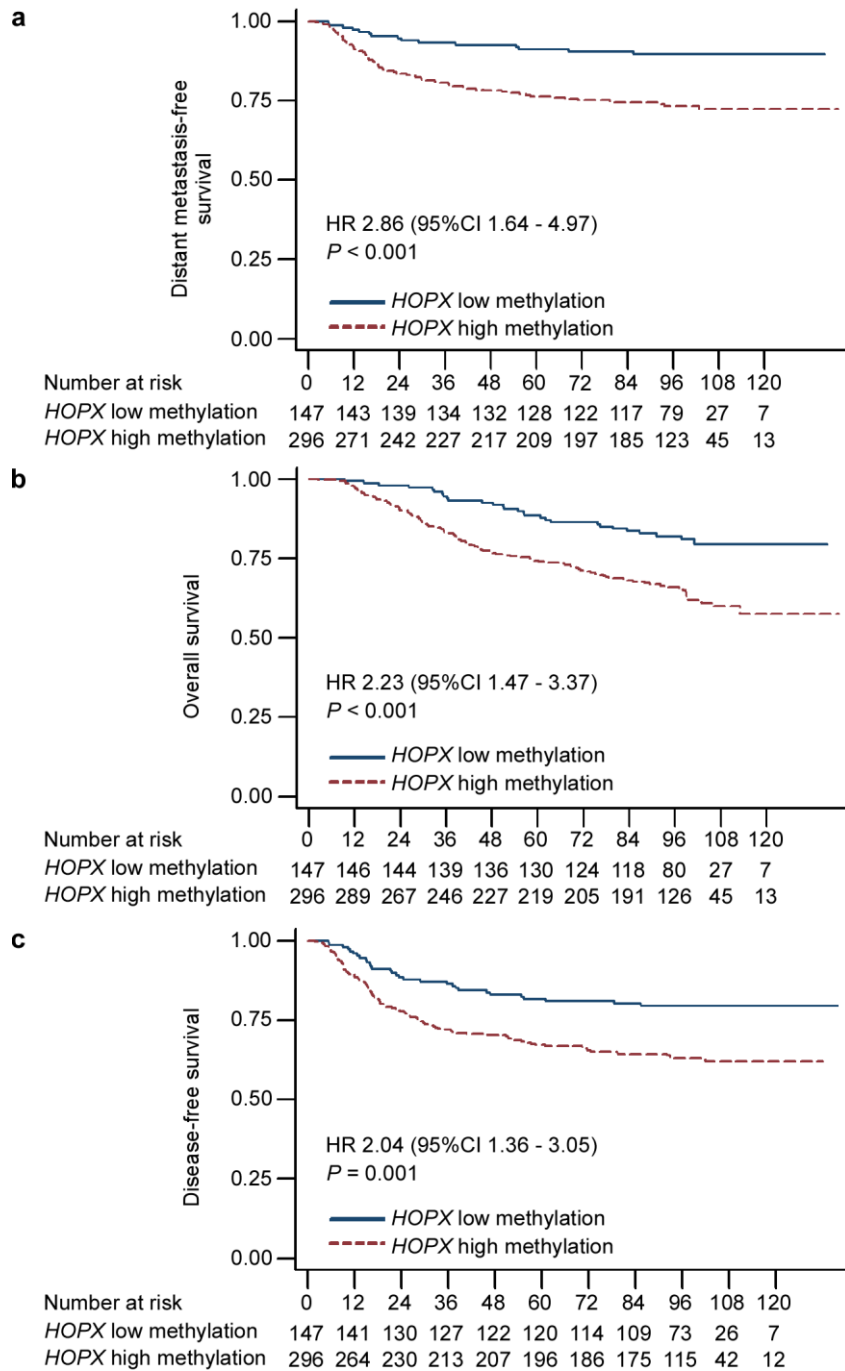


**Supplementary Figure 11. Restoring HOPX expression inhibits NPC cell aggressiveness *in vivo*.** (a) Quantification of the average volumes of the primary foot pad tumors. Mean  $\pm$  s.d.;  $P > 0.05$  compared with vector; Student's *t*-tests. (b) Representative images ( $200\times$ ) of the microscopic primary tumor in foot pad stained with H & E. Arrows represent lymphatic vessels. Scale bar:  $100\ \mu\text{m}$ . These data are representative of eight independent experiments (each mouse sample was considered as one independent experiment; three technological replications were repeated in each sample).



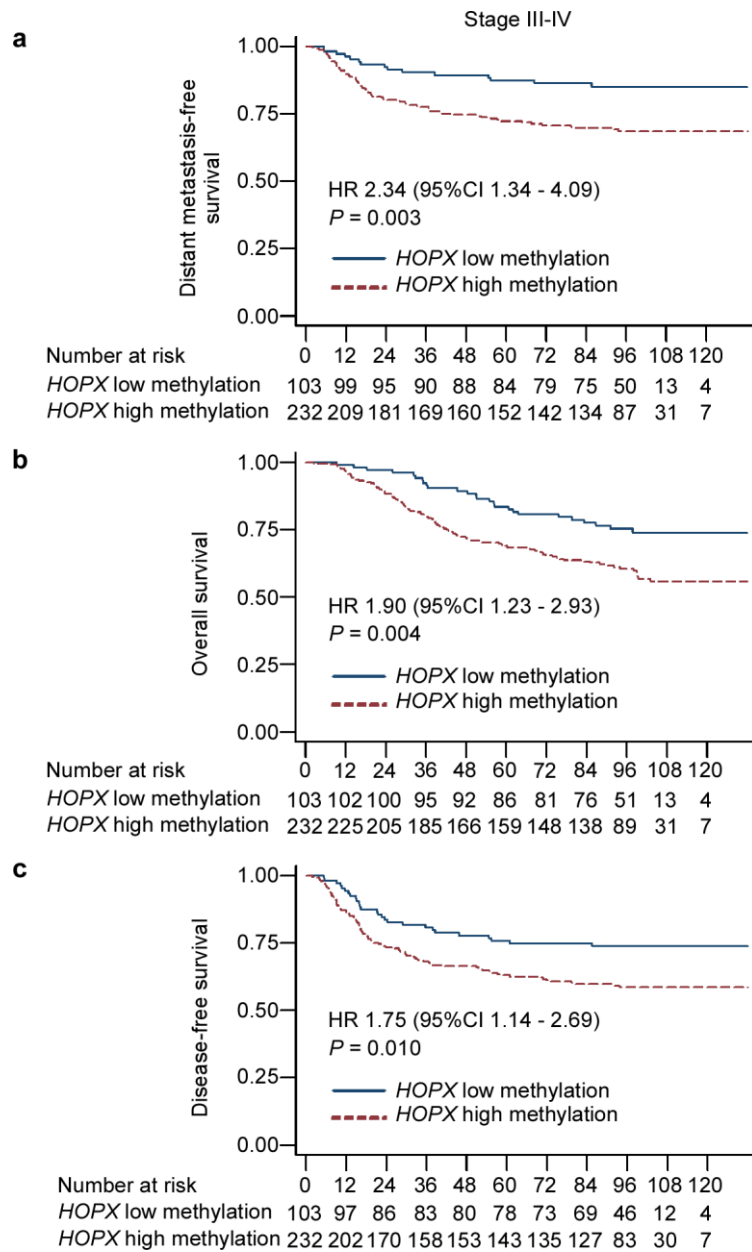
**Supplementary Figure 12. The methylation level of *HOPX* is positively associated with the *SNAIL* mRNA level in NPC. (a) *HOPX* methylation levels were determined via bisulfite pyrosequencing, while the *SNAIL* mRNA levels were determined via real time RT-PCR in NPC tissues (n = 24). Statistical analysis was performed using the Pearson's coefficient test. These data are representative of three independent experiments.**



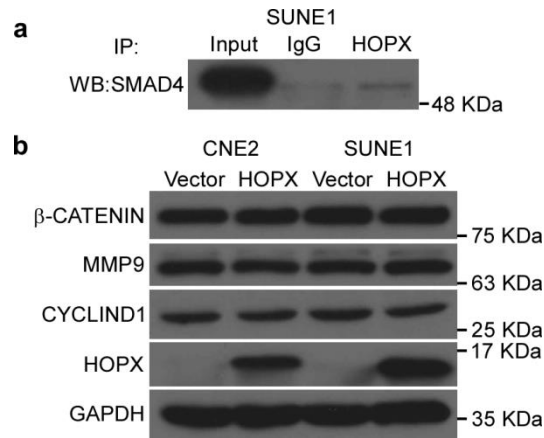


**Supplementary Figure 13. *HOPX* hypermethylation is associated with poor prognosis in the combined cohort of NPC.** All patients in the training and validation cohorts were combined (n = 443). (a-c) Kaplan-Meier analysis was performed to determine the DMFS (a), OS (b) and DFS (c) according to *HOPX* methylation levels (low methylation vs high methylation) in NPC patients. Adjusted univariate Cox proportional hazard models were used to calculate the HR values and *P* values.





**Supplementary Figure 14. HOPX hypermethylation is associated with poor prognosis stratified by TNM stage. (a) DMFS, (b) OS and (c) DFS in NPC patients within TNM stage III-IV (n = 335). Adjusted univariate Cox proportional hazard models were used to calculate the HR values and P values.**



**Supplementary Figure 15. HOPX exhibits little effect on the WNT signaling pathway. (a)** Co-IP assay was used to measure the interaction between HOPX and SMAD4 in SUNE1 cells with stable HOPX overexpression. **(b)** Western blotting assay of HOPX,  $\beta$ -CATENIN, MMP9 and CYCLIND1 expression in CNE2 and SUNE1 cells with vector or HOPX overexpression. These data are representative of three independent experiments.

**Supplementary Table 1.** The top 10 differentially expressed genes between SUNE1 cells with HOPX or vector overexpression using tumor metastasis PCR array. *P* value is compared with the control using Student's *t*-tests; genes are ranked by *P* value. These data are representative of three independent experiments.

Gene	Functional Gene Grouping	Fold change	P value
<i>CXCL12</i>	Cytokines	3.26595684	0.000110047
<i>IL1B</i>	Cell Cycle Regulation; Negative Regulation of Cell Proliferation; Cytokines; Apoptosis	48.6101703	0.000908618
<i>NME4</i>	Other Genes Related to Metastasis	37.14376425	0.001387345
<i>MMP2</i>	Matrix Metalloproteinases	21.40828154	0.001792716
<i>EPHB2</i>	Receptors	3.133358068	0.002630822
<i>SERPINE1</i>	Other ECM Proteins	10.77413353	0.003052426
<i>COL4A2</i>	Other ECM Proteins	4.537641016	0.004445524
<b><i>ECADHERIN</i></b>	<b>Cell to Cell Adhesion</b>	<b>2.213765777</b>	<b>0.004685254</b>
<i>SYK</i>	Cell to Cell Adhesion; Other Genes Related to Growth	0.265441999	0.005936684
<i>CD44</i>	Cell to Cell Adhesion; Transmembrane Receptors	3.385831498	0.006076176

**Supplementary Table 2.** Clinical characteristics of Nasopharyngeal Carcinoma patients according to the *HOPX* methylation level in the training and validation Cohorts.

Characteristic	Training Cohort (n = 255)			Validation Cohort (n = 188)		
	Low	High	<i>P</i> value *	Low	High	<i>P</i> value *
	Methylation Group (%)	Methylation Group (%)		Methylation Group (%)	Methylation Group (%)	
	n=70	n=185	n=77	n=111		
Age (mean ± S.D.)						
< 45	41 (58.6)	93 (50.3)	0.236	35 (45.5)	37 (33.3)	0.093
≥ 45	29 (41.4)	92 (49.7)		42 (54.5)	74 (66.7)	
Sex						
Female	17 (24.3)	52(28.1)	0.540	23 (29.9)	29 (26.1)	0.573
Male	53 (75.7)	133(71.9)		54 (70.1)	82 (73.9)	
WHO pathologic type						
I + II	1 (1.4)	5 (2.7)	0.475	8 (10.4)	6 (5.4)	0.201
III	69 (98.6)	180(97.3)		69(89.6)	105 (94.6)	
VCA-IgG						
< 80	9 (12.9)	20(10.8)	0.646	55 (71.4)	65 (58.6)	0.071
≥ 80	61 (87.2)	165(89.2)		22 (28.6)	46 (41.4)	
EA-IgG						
< 10	16 (22.9)	33(17.8)	0.364	24 (31.2)	24 (21.6)	0.104
≥ 10	54 (77.2)	152(82.2)		53 (68.8)	87 (78.4)	
TNM Stage						
I-II	23 (32.9)	38 (20.5)	<b>0.040</b>	21(27.3)	26 (23.4)	0.549
III-IV	47 (67.1)	147 (79.5)		56 (72.7)	85(76.6)	
Relapses or deaths						
No	57 (81.4)	120 (64.9)	<b>0.010</b>	60 (77.9)	67 (60.4)	<b>0.011</b>
Yes	13 (18.6)	65 (35.1)		17 (22.1)	44 (39.6)	

Distant metastasis

No	63 (90.0)	137 (74.1)	<b>0.006</b>	69 (89.6)	82 (73.9)	<b>0.008</b>
Yes	7 (10.0)	48 (25.9)		8 (10.4)	29 (26.1)	

Death

No	56 (80.0)	114 (61.6)	<b>0.011</b>	63 (81.8)	73 (65.8)	<b>0.016</b>
Yes	14 (20.0)	71 (38.4)		14 (18.2)	38 (34.2)	

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\* $\chi^2$  test or Fisher's exact test; TNM, tumor-node-metastasis; VCA-IgA, viral capsid antigen immunoglobulin A; EA-IgA, early antigen immunoglobulin A; All patients were restaged according to the 7<sup>th</sup> edition of the AJCC Cancer Staging Manual.



**Supplementary Table 3.** Univariate and multivariate Cox regression analysis of *HOPX* methylation level and survival.

Variable	Univariate analysis			Multivariate analysis		
	HR	95%CI	<i>P</i> value	HR	95%CI	<i>P</i> value
<b>Distant metastasis-free survival</b>						
<b>Training set</b>						
<i>HOPX</i> methylation						
(Low vs High)	2.93	1.33-6.48	<b>0.008</b>	2.44	1.10-5.43	<b>0.028</b>
TNM stage (I-II vs III-IV)	6.43	2.01-20.59	<b>0.002</b>	5.00	1.55-16.10	<b>0.007</b>
Sex (Female vs Man)	2.05	1.00-4.19	0.049			
Age ( $\leq 45$ vs $> 45$ years)	3.09	1.72-5.53	$<0.001$	2.60	1.45-4.67	0.001
WHO type (I + II vs III)	0.23	0.08-0.63	0.004	0.30	0.11-0.86	0.025
VCA-IgG ( $< 80$ vs $\geq 80$ )	2.46	0.77-7.88	0.129			
EA-IgG ( $< 10$ vs $\geq 10$ )	1.76	0.80-3.90	0.161			
<b>Validation set</b>						
<i>HOPX</i> methylation						
(Low vs High)	2.82	1.29-6.17	<b>0.009</b>	2.75	1.26-6.03	<b>0.011</b>
TNM stage (I-II vs III-IV)	3.11	1.10-8.79	<b>0.032</b>	3.02	1.07-8.52	<b>0.037</b>
Sex (Female vs Man)	1.72	0.75-3.91	0.198			
Age ( $\leq 45$ vs $> 45$ years)	2.02	0.95-4.27	0.068			
WHO type (I + II vs III)	1.41	0.34-5.85	0.639			
VCA-IgG ( $< 80$ vs $\geq 80$ )	0.70	0.35-1.43	0.329			
EA-IgG ( $< 10$ vs $\geq 10$ )	0.72	0.36-1.44	0.357			
<b>Overall survival</b>						
<b>Training set</b>						
<i>HOPX</i> methylation						
(Low vs High)	2.25	1.27-3.99	<b>0.006</b>	2.01	1.13-3.57	<b>0.018</b>
TNM stage (I-II vs III-IV)	2.98	1.54-5.77	<b>0.001</b>	2.50	1.29-4.86	<b>0.007</b>
Sex (Female vs Man)	1.95	1.12-3.42	0.019	1.87	1.07-3.28	0.028

Age ( $\leq 45$ vs $> 45$ years)	2.84	1.80-4.49	<0.001	2.58	1.63-4.08	<0.001
WHO type (I + II vs III)	0.33	0.12-0.92	0.033			
VCA-IgG ( $< 80$ vs $\geq 80$ )	2.41	0.97-5.94	0.057			
EA-IgG ( $< 10$ vs $\geq 10$ )	1.64	0.89-3.02	0.112			
<b>Validation set</b>						
<i>HOPX</i> methylation						
(Low vs High)	2.17	1.18-4.01	<b>0.013</b>	2.13	1.15-3.93	<b>0.016</b>
TNM stage (I-II vs III-IV)	3.68	1.46-9.26	<b>0.006</b>	3.61	1.43-9.08	<b>0.006</b>
Sex (Female vs Man)	1.34	0.70-2.55	0.379			
Age ( $\leq 45$ vs $> 45$ years)	1.34	0.75-2.39	0.327			
WHO type (I + II vs III)	0.98	0.35-2.71	0.966			
VCA-IgG ( $< 80$ vs $\geq 80$ )	0.81	0.45-1.45	0.475			
EA-IgG ( $< 10$ vs $\geq 10$ )	0.97	0.53-1.79	0.924			
<b>Disease-free survival</b>						
<b>Training set</b>						
<i>HOPX</i> methylation						
(Low vs High)	2.15	1.19-3.91	<b>0.012</b>	1.99	1.10-3.63	<b>0.024</b>
TNM stage (I-II vs III-IV)	2.55	1.31-4.95	<b>0.006</b>	2.16	1.11-4.22	<b>0.023</b>
Sex (Female vs Man)	1.68	0.96-2.96	0.070			
Age ( $\leq 45$ vs $> 45$ years)	2.28	1.44-3.63	<0.001	2.10	1.32-3.35	0.002
WHO type (I + II vs III)	0.33	0.12-0.88	0.027			
VCA-IgG ( $< 80$ vs $\geq 80$ )	2.14	0.87-5.30	0.099			
EA-IgG ( $< 10$ vs $\geq 10$ )	1.30	0.72-2.36	0.386			
<b>Validation set</b>						
<i>HOPX</i> methylation						
(Low vs High)	2.03	1.16-3.55	<b>0.013</b>	1.96	1.12-3.43	<b>0.019</b>
TNM stage (I-II vs III-IV)	3.06	1.39-6.73	<b>0.005</b>	2.97	1.35-6.53	<b>0.007</b>
Sex (Female vs Man)	1.49	0.81-2.75	0.202			
Age ( $\leq 45$ vs $> 45$ years)	1.62	0.93-2.81	0.086			

WHO type (I + II vs III)	1.63	0.51-5.19	0.411
VCA-IgG (< 80 vs ≥ 80)	0.70	0.35-1.43	0.329
EA-IgG (< 10 vs ≥ 10)	0.89	0.51-1.56	0.679

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Adjusted multivariate Cox proportional hazard models were used to calculate the HR values and P values, including *HOPX* methylation level (low vs high), TNM stage (stage I-II vs III-IV), sex (female vs male), age (< 45 vs ≥ 45 years), WHO pathological type (I + II vs III), VCA-IgA (< 1:80 vs ≥ 1:80) and EA-IgA (< 1:10 vs ≥ 1:10). Variables which were significantly associated with survival are presented.

**Supplementary Table 4.** Multivariate Cox regression analysis of *HOPX* methylation level and survival in the combined cohort.

Variable	HR	95%CI	P value
<b>Distant metastasis-free survival</b>			
<i>HOPX</i> methylation (Low vs High)	2.52	1.45-4.38	<b>0.001</b>
TNM stage (I-II vs III-IV)	3.83	1.77-8.31	<b>0.001</b>
Age ( $\leq 45$ vs $> 45$ years)	2.30	1.45-3.65	<b>&lt;0.001</b>
<b>Overall survival</b>			
<i>HOPX</i> methylation (Low vs High)	2.03	1.34-3.08	<b>0.001</b>
TNM stage (I-II vs III-IV)	2.80	1.64-4.81	<b>&lt;0.001</b>
Age ( $\leq 45$ vs $> 45$ years)	1.95	1.36-2.80	<b>&lt;0.001</b>
<b>Disease-free survival</b>			
<i>HOPX</i> methylation (Low vs High)	1.84	1.23-2.77	<b>0.003</b>
TNM stage (I-II vs III-IV)	2.44	1.46-4.06	<b>0.001</b>
Age ( $\leq 45$ vs $> 45$ years)	1.85	1.29-2.63	<b>0.001</b>

Adjusted multivariate Cox proportional hazard models were used to calculate the HR values and P values, including *HOPX* methylation level (low vs high), TNM stage (stage I-II vs III-IV), sex (female vs male), age ( $< 45$  vs  $\geq 45$  years), WHO pathological type (I + II vs III), VCA-IgA ( $< 1:80$  vs  $\geq 1:80$ ) and EA-IgA ( $< 1:10$  vs  $\geq 1:10$ ). Variables which were significantly associated with survival are presented.

**Supplementary Table 5.** Primers used in this study.

Gene	Sequence (5' to 3')
<b>BSP PCR primers</b>	
<i>HOPX</i> -F	GTTCTTAGGGATTTTTGTAGGAATTG
<i>HOPX</i> -R	CTTCCTTCACTCCTTCCTTAAAAC (5'Biotin)
<b>BSP sequencing primer</b>	
<i>HOPX</i>	GGATTTTTGTAGGAATTGTAT
<b>Real time RT-PCR primers</b>	
<i>HOPX</i> -F	CACCACGCTGTGCCTCAT
<i>HOPX</i> -R	CCATTTCTGGGTCTCCTCC
<i>SNAIL</i> -F	TAATCCAGAGTTTACCTTCCAGC
<i>SNAIL</i> -R	CTCATCTGACAGGGAGGTCAG
<i>SLUG</i> -F	CCCTGAAGATGCATATTCGGA
<i>SLUG</i> -R	CTGCAAATGCTCTGTTGCAG
<i>ZEB1</i> -F	TCACTAGTGTTTACCAGAACAGTG
<i>ZEB1</i> -R	GAACACTGTTCTGGTCAGCA
<i>ZEB2</i> -F	CTACAAGCGCTTGACATCAC
<i>ZEB2</i> -R	TAGCATTTGGTGCTGATCTGTC
<i>TWIST1</i> -F	AGTCTTACGAGGAGCTGCAG
<i>TWIST1</i> -R	CTCTGGAGGACCTGGTAGAG
<i>FOXC2</i> -F	CTTCTACCGGGAGAACAAGC
<i>FOXC2</i> -R	CTCCTTCTCCTTGGACACGT
<i>ECADHERIN</i> -F	GAAGAGGACCAGGACTTTGAC
<i>ECADHERIN</i> -R	GTAGTCATAGTCCTGGTCTTTGTC
<i>α-CATENIN</i> -F	CCTGAGGAGTTGGATGACTC
<i>α-CATENIN</i> -R	TCCTTTACCTCGGGTAAAGTC



<i>VIMENTIN-F</i>	ATTCCACTTTGCGTTCAAGG
<i>VIMENTIN-R</i>	CTTCAGAGAGAGGAAGCCGA
<i>FIBRONECTIN-F</i>	CACGATGATATGGAGAGCCA
<i>FIBRONECTIN-R</i>	TATTTGGTGGCCACCATAAGTC
<i>SRF-F</i>	CACCATCATGACGTCATCCG
<i>SRF-R</i>	CTATCACAGCCATCTGGTGG

#### **siRNA sequences**

siRNA-NC-F	5'-UUCUCCGAACGUGUCACGUTT-3'
siRNA-NC-R	5'-ACGUGACACGUUCGGAGAATT-3'
siHOPX-1-F	5'-GAAGCUAUGUGUAUCUUCUTT-3'
siHOPX-1-R	5'-AGAAGAUACACAUAGCUUCTT-3'
siHOPX-2-F	5'-CCCUAAGUCACUUUCCUUATT-3'
siHOPX-2-R	5'-UAAGGAAAGUGACUUAGGGTT-3'

#### **ChIP-PCR primers**

<i>SNAIL-1-F</i>	AGGAGTCCCCGCCCCGGGCTCTCACC
<i>SNAIL-1-R</i>	GCAGCAGCGCCGCCAACTCCCTTAA
<i>SNAIL-2-F</i>	GGGGCGTCAGAAGCGCTCAGACCAC
<i>SNAIL-2-R</i>	GTTCGCTGGCGCAGCGCGGGTCGTC
<i>SNAIL-3-F</i>	GTGTCCCTTTCTCGCTTCCTC
<i>SNAIL-3-R</i>	CGGGACACCTGACCTTCCGAC
<i>SNAIL-4-F</i>	TTCTGTCCGGGGCTGTGCCCTG
<i>SNAIL-4-R</i>	CGCACGTGGCTCTCGGCGGCTTG
<i>SNAIL-5-F</i>	AGTCACCCCGACCCCTGTCAG
<i>SNAIL-5-R</i>	CACTCAGAGCCTCTCCCGAAG
<i>SNAIL-6-F</i>	TCCAAACTCCTACGAGGCCCTG
<i>SNAIL-6-R</i>	TCCCAGAGGAAGTGAAGAATTA

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**Supplementary Table 6.** Antibodies used in this study.

Antibody	Company	Catalog no.	Dilution
<b>Western blotting</b>			
GAPDH	Santa Cruz	sc-25778	1:1000
HOPX	Santa Cruz	SC-30216	1:200
ECADHERIN	BD	610181	1:2000
$\alpha$ -CATENIN	BD	610193	1:500
VIMENTIN	Proteintech	10366-1-AP	1:1000
FIBRONECTIN	Proteintech	156131-AP	1:500
SNAIL	Abcam	ab180714	1:1000
SRF	Proteintech	16821-1-AP	1:800
HDAC2	Proteintech	12922-3-AP	1:1000
SMAD4	Proteintech	10231-1-AP	1:200
$\beta$ -CATENIN	Proteintech	51067-2-AP	1:500
CYCLIND1	Proteintech	60186-1-Ig	1:500
MMP9	Proteintech	10375-2-AP	1:500
Mouse	CST	7076	1:5000
Rabbit	CST	7074	1:5000
<b>Immunohistochemistry</b>			
HOPX	Santa Cruz	SC-30216	1:100
SNAIL	Abcam	ab180714	1:200
<b>Immunofluorescence</b>			
SRF	Proteintech	16821-1-AP	1:100
HDAC2	Santa Cruz	SC-6296	1:100
HOPX	Santa Cruz	SC-30216	1:100
ECADHERIN	BD	610181	1:100
$\alpha$ -CATENIN	BD	610193	1:100

VIMENTIN	Proteintech	10366-1-AP	1:100
goat Anti-rabbit, Alexa Fluor® 594 IgG secondary antibody	Life	A-11008	1:1000
goat Anti-mouse, Alexa Fluor® 594 IgG secondary antibody	Life	A-11001	1:1000
rabbit Anti-goat, Alexa Fluor® 647 IgG secondary antibody	Life	A-27018	1:1000

### **Co-Immunoprecipitation**

HOPX	Santa Cruz	SC-30216	3µg
SRF	Proteintech	16821-1-AP	3µg
HDAC2	Proteintech	12922-3-AP	3µg
IgG	Proteintech	30000-0-AP	3µg

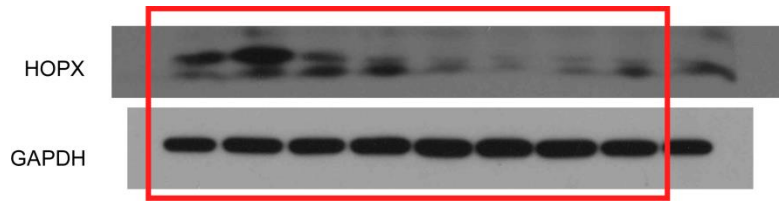
### **Chromatin Immunoprecipitation**

HOPX	Santa Cruz	SC-30216	2.0 ml per 0.5 ml Protein A/G sepharose beads
HDAC2	Abcam	ab7029	0.5mg per 5ml Protein A/G sepharose beads
H3K9Ac	Abcam	ab10812	0.2mg per 2ml Protein A/G sepharose beads
SRF	CST	5147	1:100
IgG	Sigma	SLBJ3775V	1:100

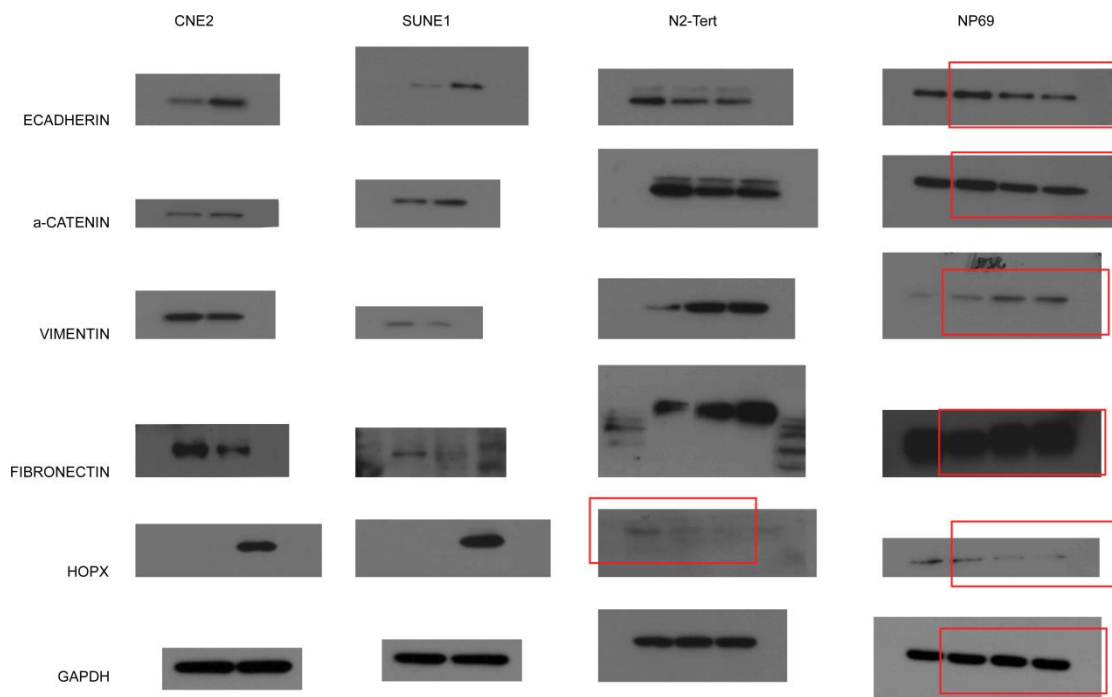
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**Supplementary Figure 16.** Full unedited western blotting gels for all figures.

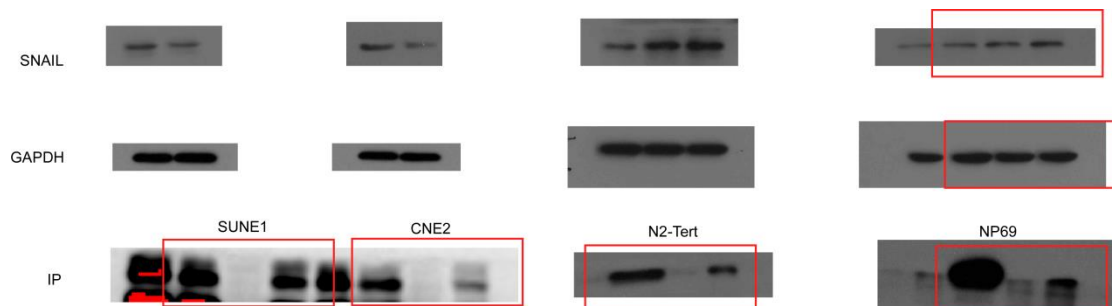
Full unedited gel for Figure 2e



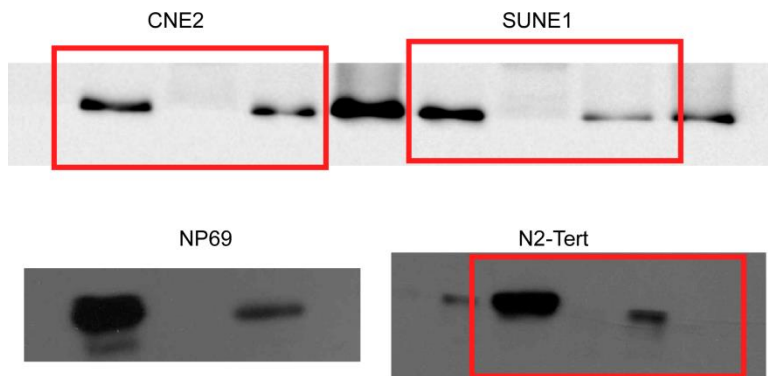
Full unedited gel for Figure 4d,e



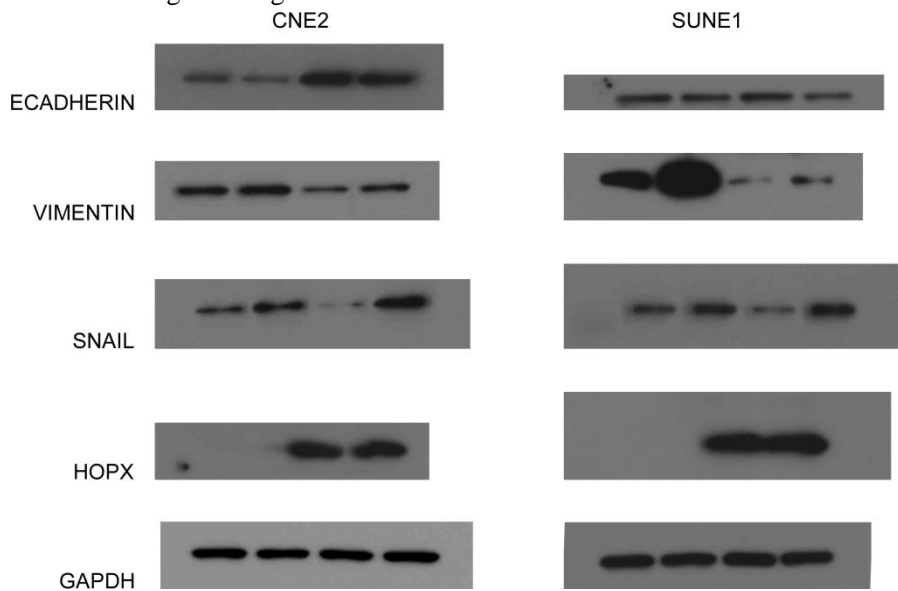
Full unedited gel for Figure 5c, d and g



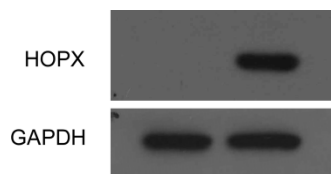
Full unedited gel for Figure 6c



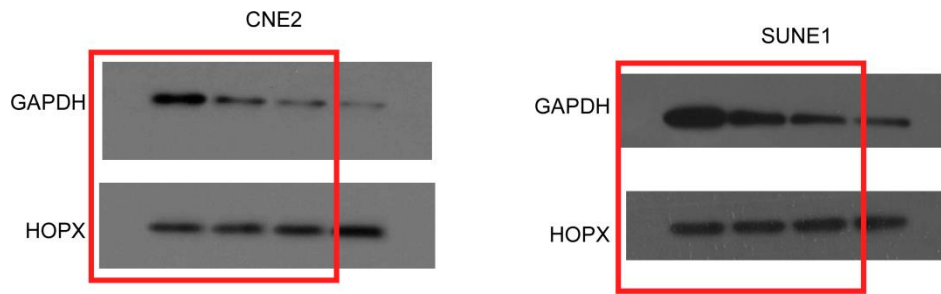
Full unedited gel for Figure 7d



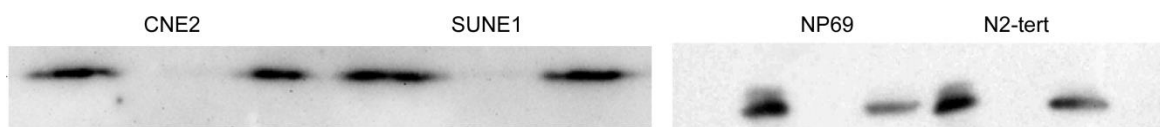
Full unedited gel for Supplementary Figure 3a



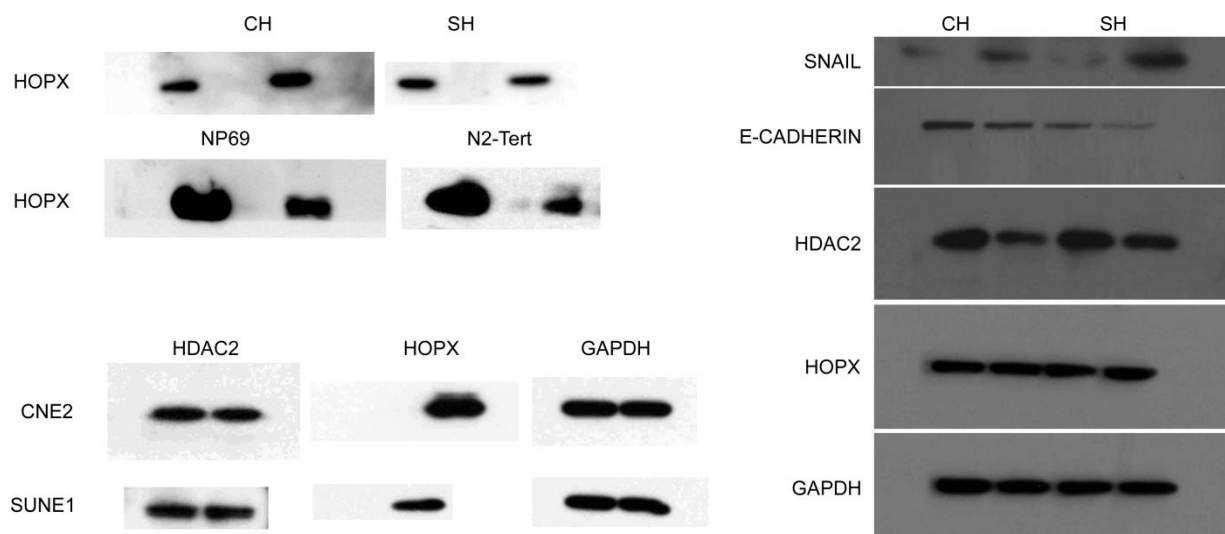
Full unedited gel for Supplementary Figure 4a



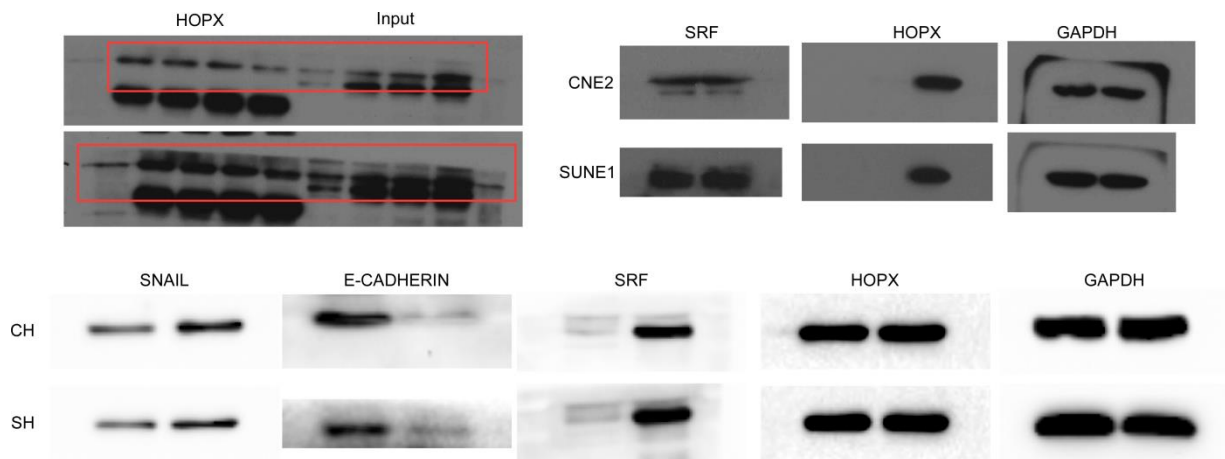
Full unedited gel for Supplementary Figure 7c



Full unedited gel for Supplementary Figure 8a,b,d



Full unedited gel for Supplementary Figure 9b,c,g



Full unedited gel for Supplementary Figure 15

