

Figure S1. Targeting MUC1-C downregulates BRG1 and ARID1A expression in different types of carcinoma cells. A. Lysates from DU-145 cells stably expressing a CshRNA or MUC1shRNA were immunoblotted with antibodies against the indicated proteins. B and C. BT-549 TNBC (B) and SW620 colon cancer (C) cells expressing a control tet-CshRNA or tet-MUC1shRNA were treated with vehicle or DOX for 7 days. Lysates were immunoblotted with antibodies against the indicated proteins.

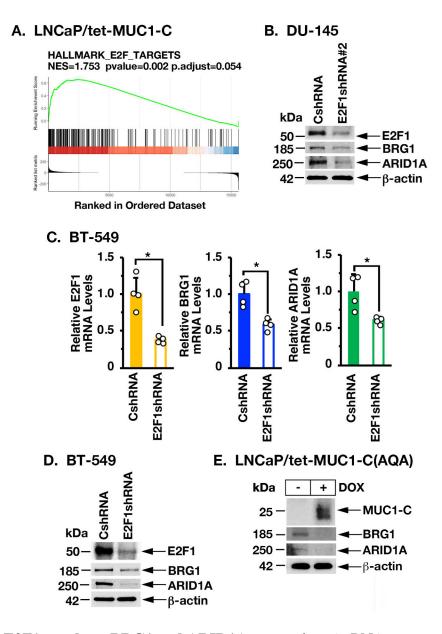


Figure S2. E2F1 regulates BRG1 and ARID1A expression. A. RNA-seq was performed in triplicate on LNCaP/tet-MUC1-C cells treated with vehicle or DOX for 7 days. The datasets were analyzed with GSEA using the Hallmark gene signature collection for E2F Targets. B. Lysates from DU-145 cells expressing a second E2F1shRNA#2 were immunoblotted with antibodies against the indicated proteins. C. BT-549/CshRNA and BT-549/E2F1shRNA cells were analyzed for the indicated mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that in control cells (assigned a value of 1). D. Lysates from BT-549/CshRNA and BT-549/E2F1shRNA cells were immunoblotted with antibodies against the indicated proteins. E. Lysates from LNCaP/tet-MUC1-C(AQA) cells treated with vehicle or DOX for 7 days were immunoblotted with antibodies against the indicated proteins.

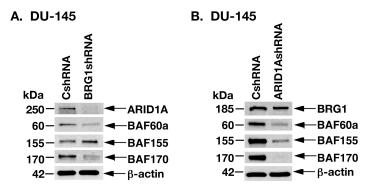


Figure S3. Effects of silencing BRG1 and ARID1A on expression of other BAF subunits. A and B. Lysates from DU-145 cells expressing a CshRNA and BRG1shRNA (A) or ARID1AshRNA (B) were immunoblotted with antibodies against the indicated proteins.

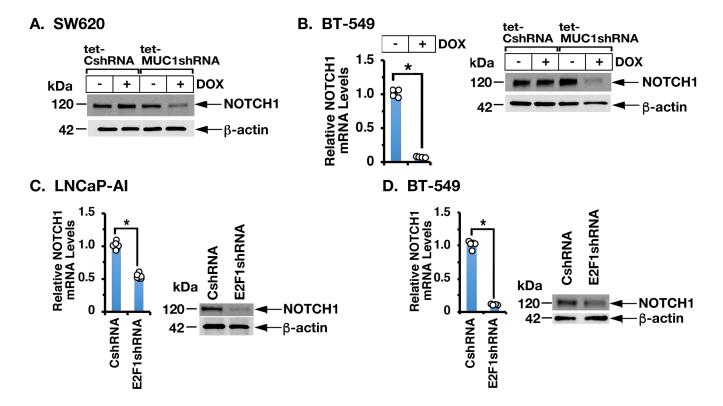


Figure S4. Silencing MUC1-C and E2F1 downregulates NOTCH1 in carcinoma cells. A. SW620/tet-CshRNA and SW620/tet-MUC1shRNA cell were treated with vehicle or DOX for 7 days. Lysates were immunoblotted with antibodies against the indicated proteins. B. BT-549/tet-MUC1shRNA cells treated with vehicle or DOX for 7 days were analyzed for NOTCH1 mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that in control cells (assigned a value of 1) (left). Lysates from BT-549/tet-CshRNA and BT-549/tet-MUC1shRNA cells treated with vehicle or DOX for 7 days were immunoblotted with antibodies against the indicated proteins (right). C and D. LNCaP-AI (C) and BT-549 (D) cells expressing a CshRNA or E2F1shRNA were analyzed for NOTCH1 mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that in control cells (assigned a value of 1) (left). Lysates were immunoblotted with antibodies against the indicated proteins (right).

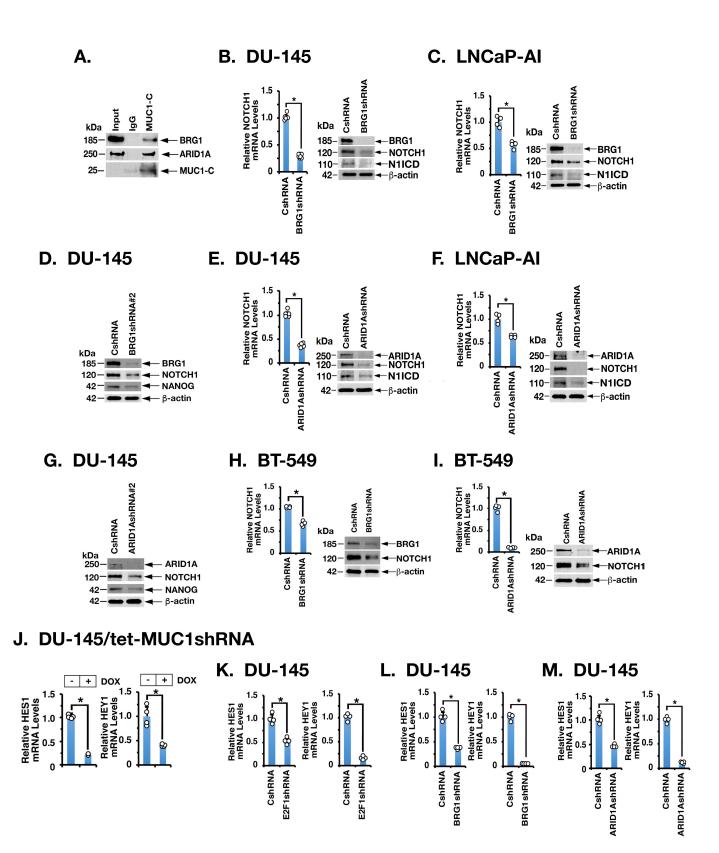


Figure S5. Effects of targeting the MUC1-C→E2F1→BAF pathway on NOTCH1, HES1 and HEY1 expression. A. Nuclear lysates from DU-145 cells were incubated with a control IgG and anti-MUC1-C. The input lysate and precipitates were immunoblotted with antibodies against the indicated proteins. B and C. DU-145 (B) and LNCaP-AI (C) cells expressing a CshRNA or BRG1shRNA were analyzed for NOTCH1 mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are

expressed as relative mRNA levels compared to that in control cells (assigned a value of 1) (left). Lysates were immunoblotted with antibodies against the indicated proteins (right). D. Lysates from DU-145 cells expressing a CshRNA and a second BRG1shRNA#2 were immunoblotted with antibodies against the indicated proteins. E and F. DU-145 (E) and LNCaP-AI (F) cells expressing a CshRNA or ARID1AshRNA were analyzed for NOTCH1 mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that in control cells (assigned a value of 1) (left). Lysates were immunoblotted with antibodies against the indicated proteins (right). G. Lysates from DU-145 cells expressing a CshRNA and a second ARID1AshRNA#2 were immunoblotted with antibodies against the indicated proteins. H and I. BT-549/CshRNA, BT-549/BRG1shRNA (H) and BT-549/ARID1AshRNA (I) cells were analyzed for NOTCH1 mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that in control cells (assigned a value of 1) (left). Lysates were immunoblotted with antibodies against the indicated proteins (right). J. DU-145/tet-MUC1shRNA cells treated with vehicle or DOX for 7 days were analyzed for the indicated mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that in control cells (assigned a value of 1). K. DU-145/CshRNA and DU-145/E2F1shRNA cells were analyzed for the indicated mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that in control cells (assigned a value of 1). L and M. DU-145/CshRNA, DU-145/BRG1shRNA (L) and DU-145/ARID1AshRNA (M) cells were analyzed for the indicated mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that in control cells (assigned a value of 1).

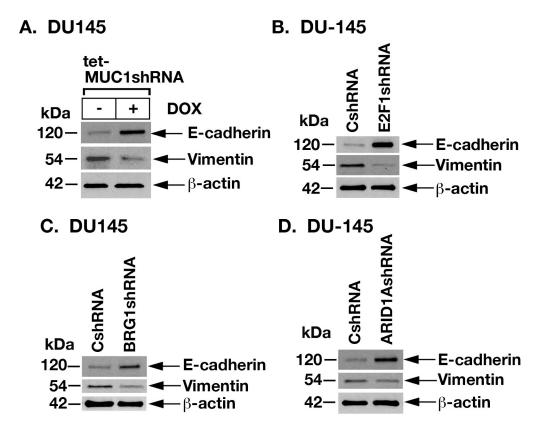


Figure S6. Targeting the MUC1-C→E2F1→BAF pathway induces E-cadherin and suppresses vimentin expression. A. Lysates from DU-145/tet-MUC1shRNA cells treated with vehicle or DOX for 7 days were immunoblotted with antibodies against the indicated proteins. B-D. Lysates from DU-145/CshRNA, DU-145/E2F1shRNA (B), DU-145/BRG1shRNA (C) and DU-145/ARID1A1shRNA (D) cells were immunoblotted with antibodies against the indicated proteins.

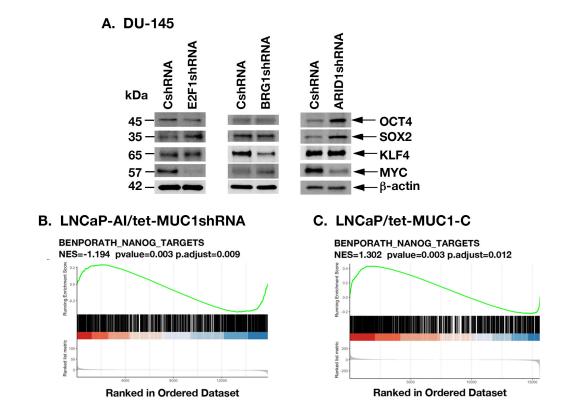


Figure S7. Effects of the MUC1-C→E2F1®→BAF pathway on OSKM expression and the NANOG TARGETS gene signature. A. Lysates from DU-145 cells expressing a CshRNA, E2F1shRNA (left), BRG1shRNA (middle) or ARID1AshRNA (right) were immunoblotted with antibodies against the indicated proteins. B and C. RNA-seq was performed in triplicate on LNCaP-AI/tet-MUC1shRNA (B) and LNCaP-AI/tet-MUC1-C (C) cells treated with vehicle or DOX for 7 days. The datasets were analyzed with GSEA using the BENPORATH_NANOG_TARGETS gene signature.

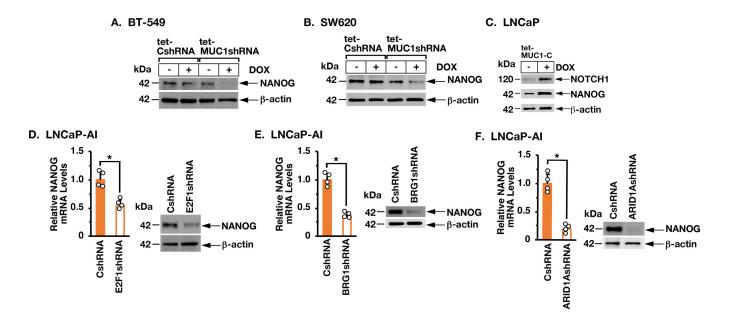


Figure S8. Silencing MUC1-C downregulates NANOG in carcinoma cells. A and B. BT-549 (A) and SW620 (B) cells expressing tet-CshRNA or tet-MUC1shRNA were treated with vehicle or DOX for 7 days. Lysates were immunoblotted with antibodies against the indicated proteins. C. LNCaP/tet-MUC1-C cells were treated with vehicle or DOX for 7 days. Lysates were immunoblotted with antibodies against the indicated proteins. D-F. LNCaP-AI/CshRNA, LNCaP-AI/E2F1shRNA (D), LNCaP-AI/BRG1shRNA (E) and LNCaP-AI/ARID1AshRNA (F) cells were analyzed for NANOG mRNA levels by qRT-PCR (left). The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that in control cells (assigned a value of 1). Lysates were immunoblotted with antibodies against the indicated proteins (right).

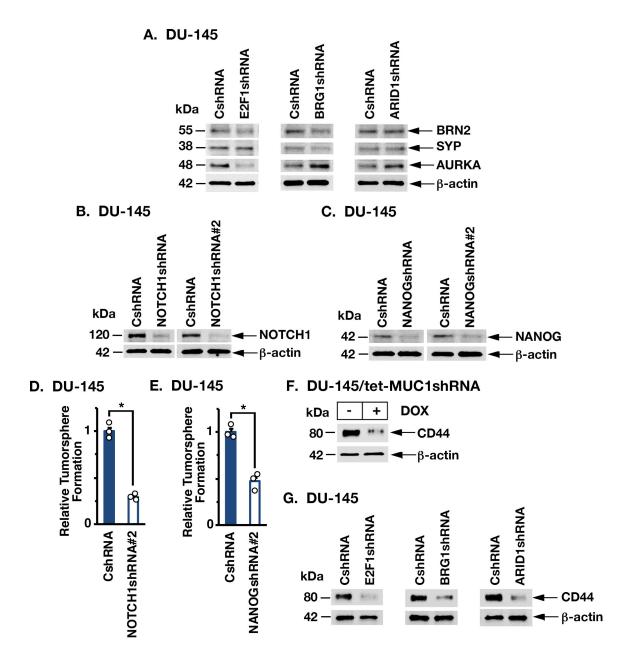


Figure S9. Silencing the MUC1-C→E2F1→BAF→NOTCH1®NANOG pathway has little effect on NE differentiation markers and suppresses CD44 expression. A. Lysates from DU-145/CshRNA, DU-145/E2F1shRNA (left), DU-145/BRG1shRNA (middle) and DU-145/ARID1A1shRNA (right) cells were immunoblotted with antibodies against the indicated proteins. B. Lysates from DU-145/CshRNA, DU-145/NOTCH1shRNA (left) and DU-145/NOTCH1shRNA#2shRNA (right) cells were immunoblotted with antibodies against the indicated proteins. C. Lysates from DU-145/CshRNA, DU-145/NANOGshRNA (left) and DU-145/NANOGshRNA#2shRNA (right) cells were immunoblotted with antibodies against the indicated proteins. D and E. DU-145 cells expressing a CshRNA, NOTCH1shRNA#2 (D) or NANOGshRNA#2 (E) were assayed for tumorsphere formation. The results (mean±SD of 3 biologic replicates) are expressed as relative tumorsphere number per field compared to the CshRNA control. F. Lysates from DU-145/tet-MUC1shRNA cells treated with vehicle or DOX for 7 days were immunoblotted with antibodies against the indicated proteins. G. Lysates from DU-145/CshRNA, DU-145/E2F1shRNA (left), DU-145/BRG1shRNA (middle) and DU-145/ARID1A1shRNA (right) cells were immunoblotted with antibodies against the indicated proteins.

Supplemental Tables

Table S1. Primers used for qRT-PCR.

MUC1-C	FWD	TACCGATCGTAGCCCCTATG					
	REV	CTCACCAGCCCAAACAGG					
BRG1	FWD	CCAAGACCCTGATGAACACC					
	REV	GGCAGAACAGCAGCACTTT					
ARID1A	FWD	ACCTCTATCGCCTCTATGTGTCTGT					
	REV	CTGGCAGCACTGCTTGATGT					
NOTCH1	FWD	GGGCTAACAAAGATATGCAG					
	REV	ACTGAACCTGACCGTACAGTTGGCAAAGTGGTCCAG					
HES1	FWD	AGATAGCTCGCGGCATTCCA					
	REV	CAGCACACTTGGGTCTGTGC					
HEY1	FWD	CATACGGCAGGAGGGAAAG					
	REV	GCATCTAGTCCTTCAATGATGCT					
NANOG	FWD	AGAGGTGAAGACCTGGTTCC					
	REV	GGTAGGTGCTGAGGCCTTCT					
GAPDH	FWD	CCATGGAGAAGGCTGGGG					
	REV	CAAAGTTGTCATGGATGACC					
	REV FWD	GGTAGGTGCTGAGGCCTTCT CCATGGAGAAGGCTGGGG					

Table S2. Primers used for ChIP-qPCR.

BRG1-Enhancer	FWD	TACGGTCCAGGGTTCCTATTT			
	REV	GGCAACTGGAGAATGGGAT			
ARID1A-Intron 1	FWD	ACATGGAAGAGGAGGAGTAT			
	REV	GGGCAAGAGTAACCTTACAGAG			
NANOG-Enhancer	FWD	CTGGGTTTGTCTTCAGGTTCT			
	REV	AATCCCGTCTACCAGTCTCA			
GAPDH	FWD	TACTAGCGGTTTTACGGGCG			
	REV	TCGAACAGGAGGAGCAGAGAGCGA			

Table S3. Stem Cell and NOTCH Datasets

Gene Set	DU-145/tet-MUC1shRNA			LNCaP-AI/tet-MUC1shRNA		
_	NES	p-val	p.adj	NES	p-val	p.adj
BHATTACHARYA_EMBRYONIC_ STEM_CELL	-1.3155	0.0711	0.3069	-2.0050	0.0021	0.0272
BOQUEST_STEM_CELL_DN	1.6940	0.0016	0.0674	0.9489	0.5664	0.7834
BOQUEST_STEM_CELL_UP	0.9270	0.6420	0.8162	1.3174	0.0454	0.1878
GO_POSITIVE_REGULATION_OF_ STEM_CELL_DIFFERENTIATION	1.0765	0.3957	0.6736	0.8429	0.6900	0.8627
GO_STEM_CELL_DIFFERENTIATION	1.5714	0.0030	0.0747	-0.8251	0.9581	0.9885
PID_NOTCH_PATHWAY	1.1345	0.2602	0.5582	1.7883	0.0121	0.0829
REACTOME_SIGNALING_BY_NOTCH	1.5408	0.0045	0.0925	1.4488	0.0128	0.0844
WONG_EMBRYONIC_STEM_CELL_CORE	-1.7093	0.0033	0.0753	-2.1443	0.0024	0.0285