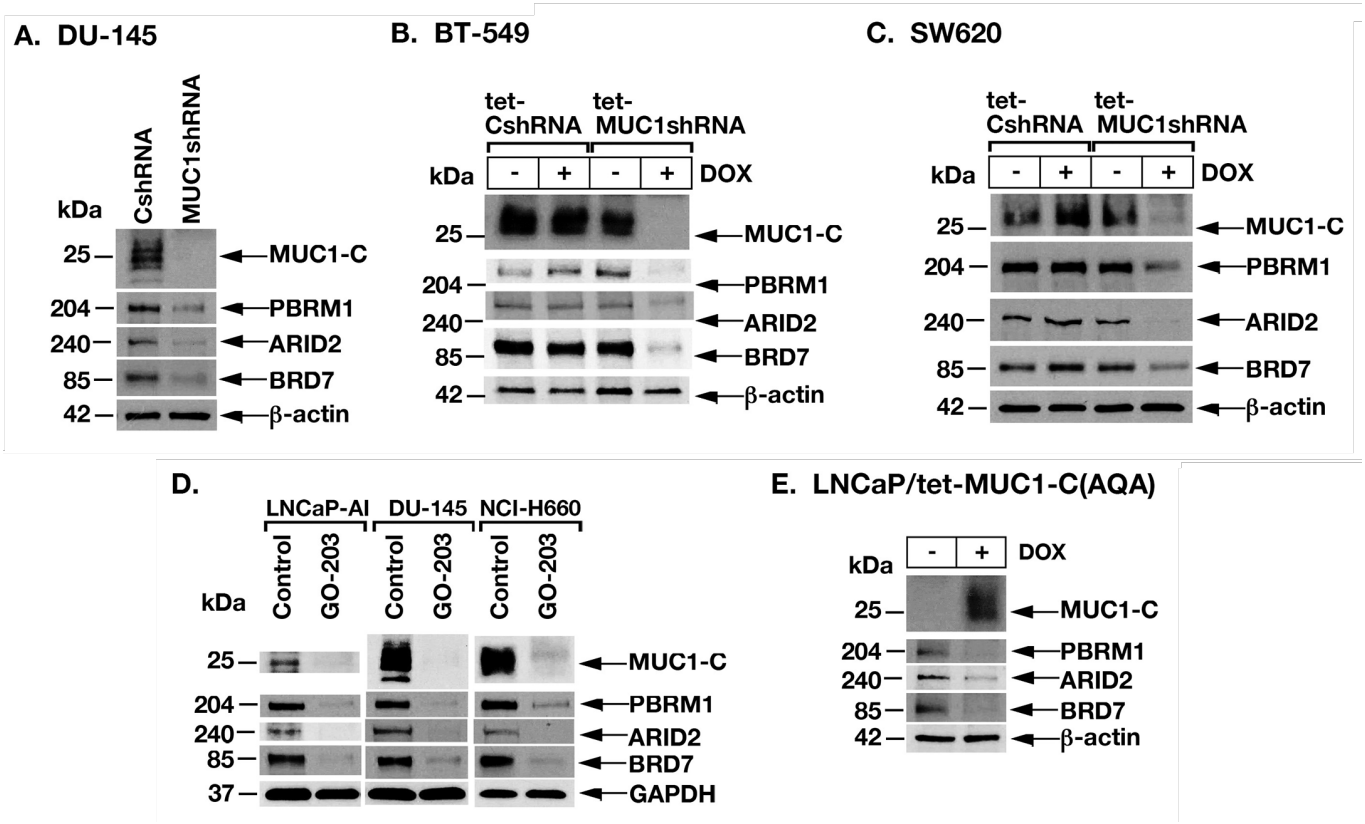
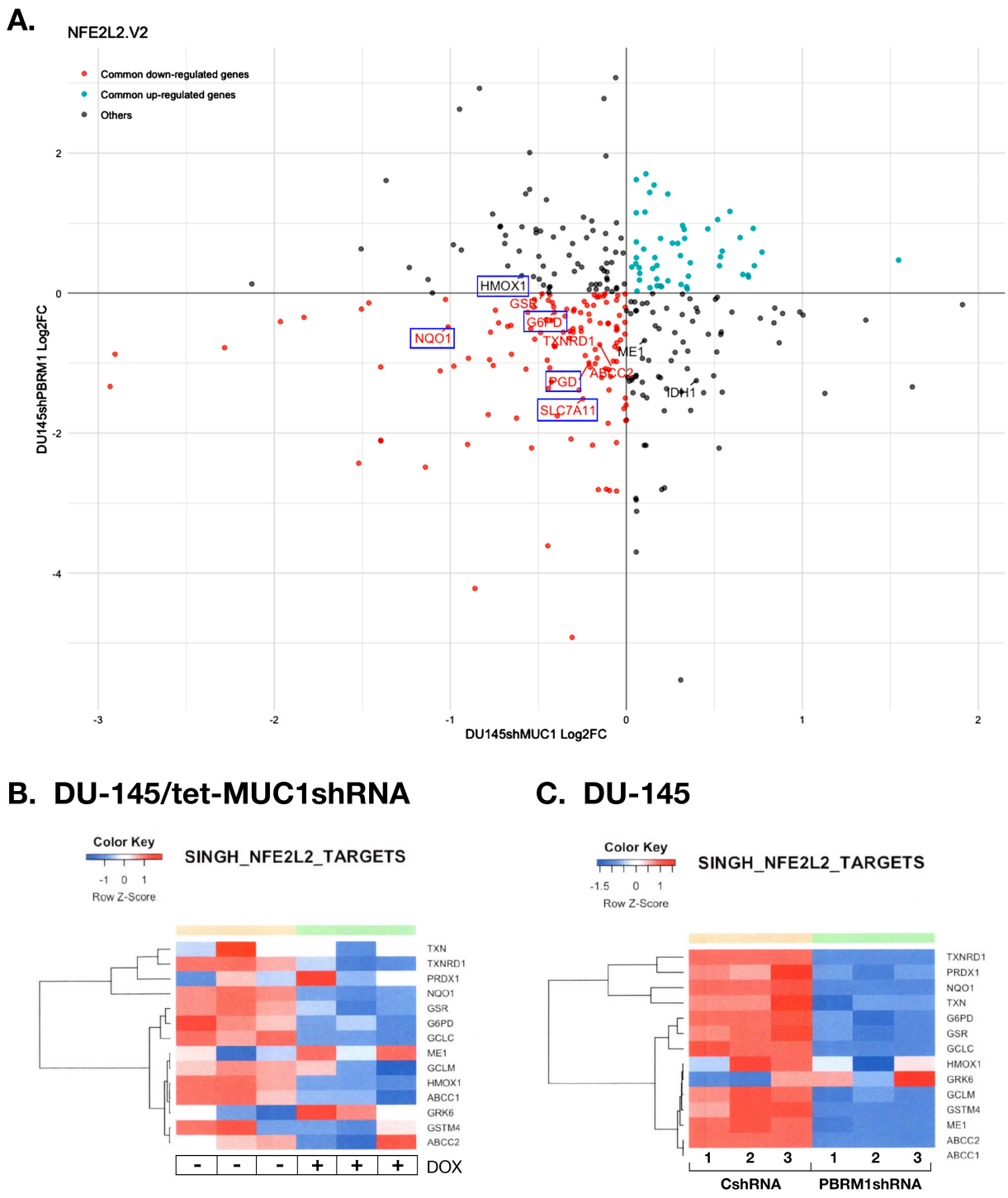
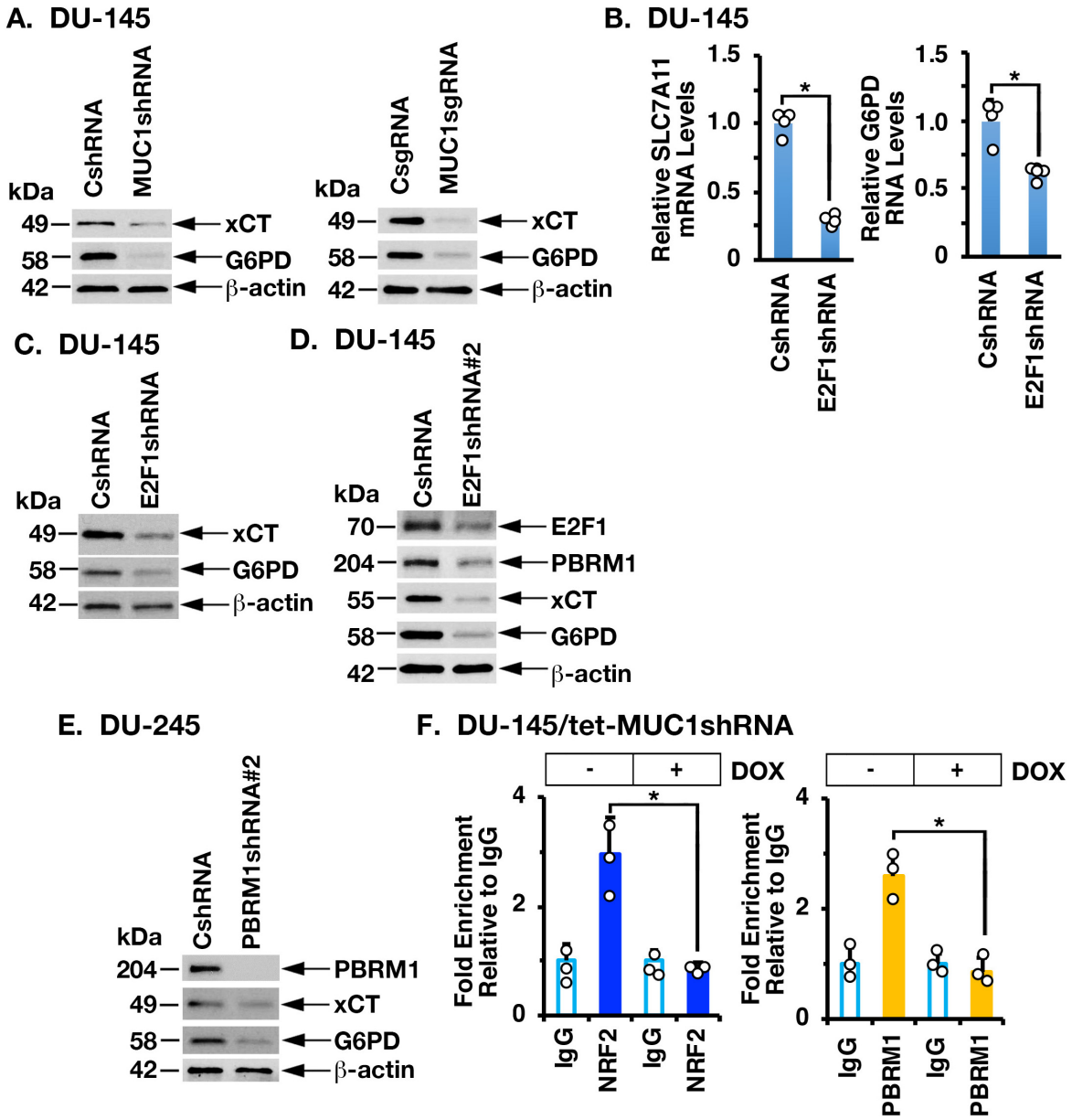


Supplemental Figure S1. Targeting MUC1-C downregulates PBRM1, ARID2 and BRD7 expression in human cancer cells. A. Lysates from DU-145/CshRNA and DU-145/MUC1shRNA cells were immunoblotted with antibodies against the indicated proteins. **B and C.** Lysates from BT-549/tet-CshRNA and BT-549/tet-MUC1shRNA (**B**) or SW620/tet-CshRNA and SW620/tet-MUC1shRNA (**C**) cells treated with vehicle or DOX for 7 days were immunoblotted with antibodies against the indicated proteins. **D.** Lysates from LNCaP-AI, DU145 and NCI-H660 cells treated with 5 μM GO-203 for 2 days 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.

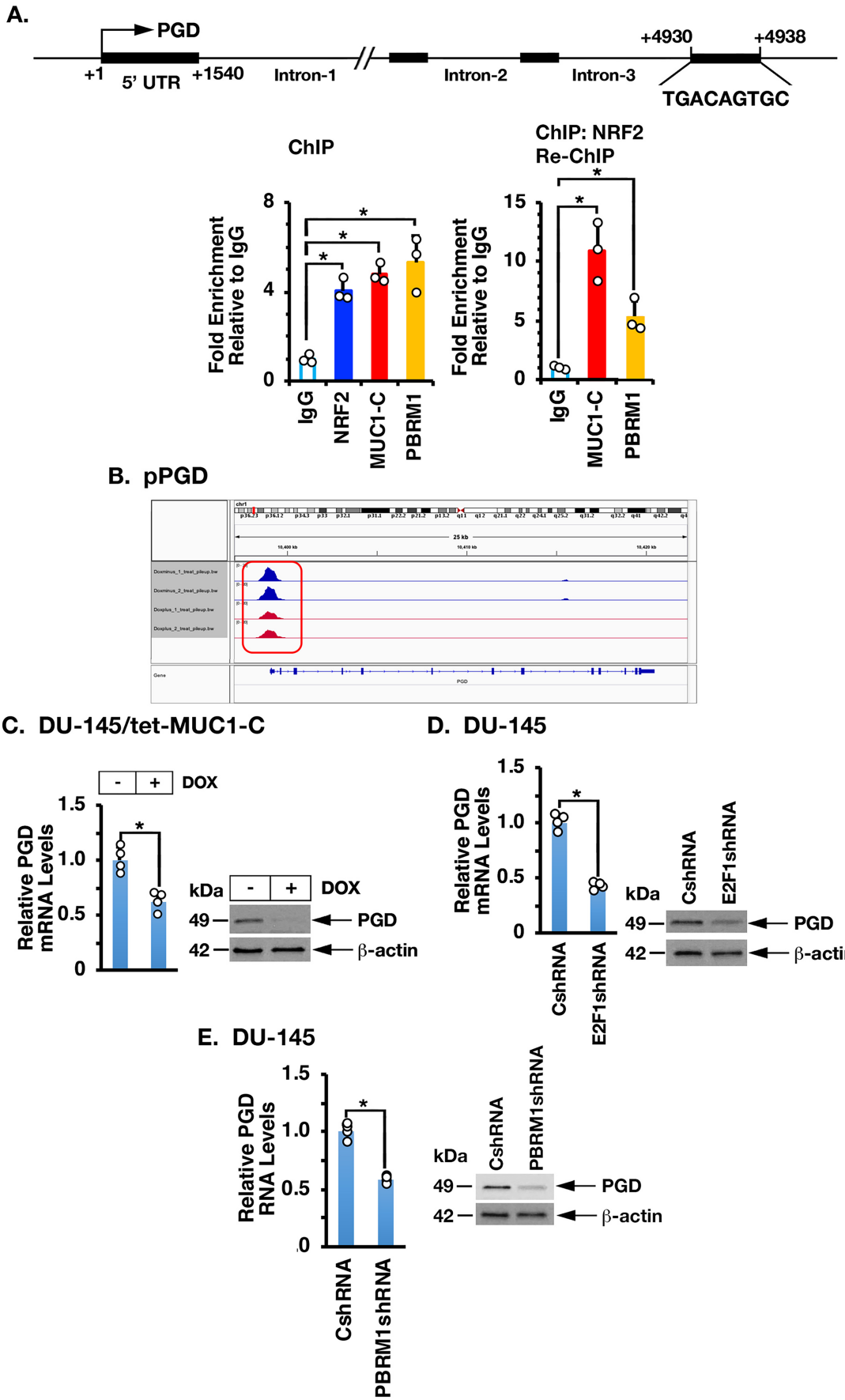


Supplemental Figure S2. MUC1-C and PBRM1 activate NRF2 gene signatures. A. Overlap of down- and up-regulated genes in DU-145 cells with MUC1-C and PBRM1 silencing obtained from GSEA of the NFE2L2.V2 gene signature. Highlighted in the blue boxes are key NRF2-induced antioxidant genes. **B and C.** Heatmaps of MUC1-C **(B)** and PBRM1 **(C)** regulated genes identified from GSEA of the SINGH NFE2L2 TARGETS gene signature.

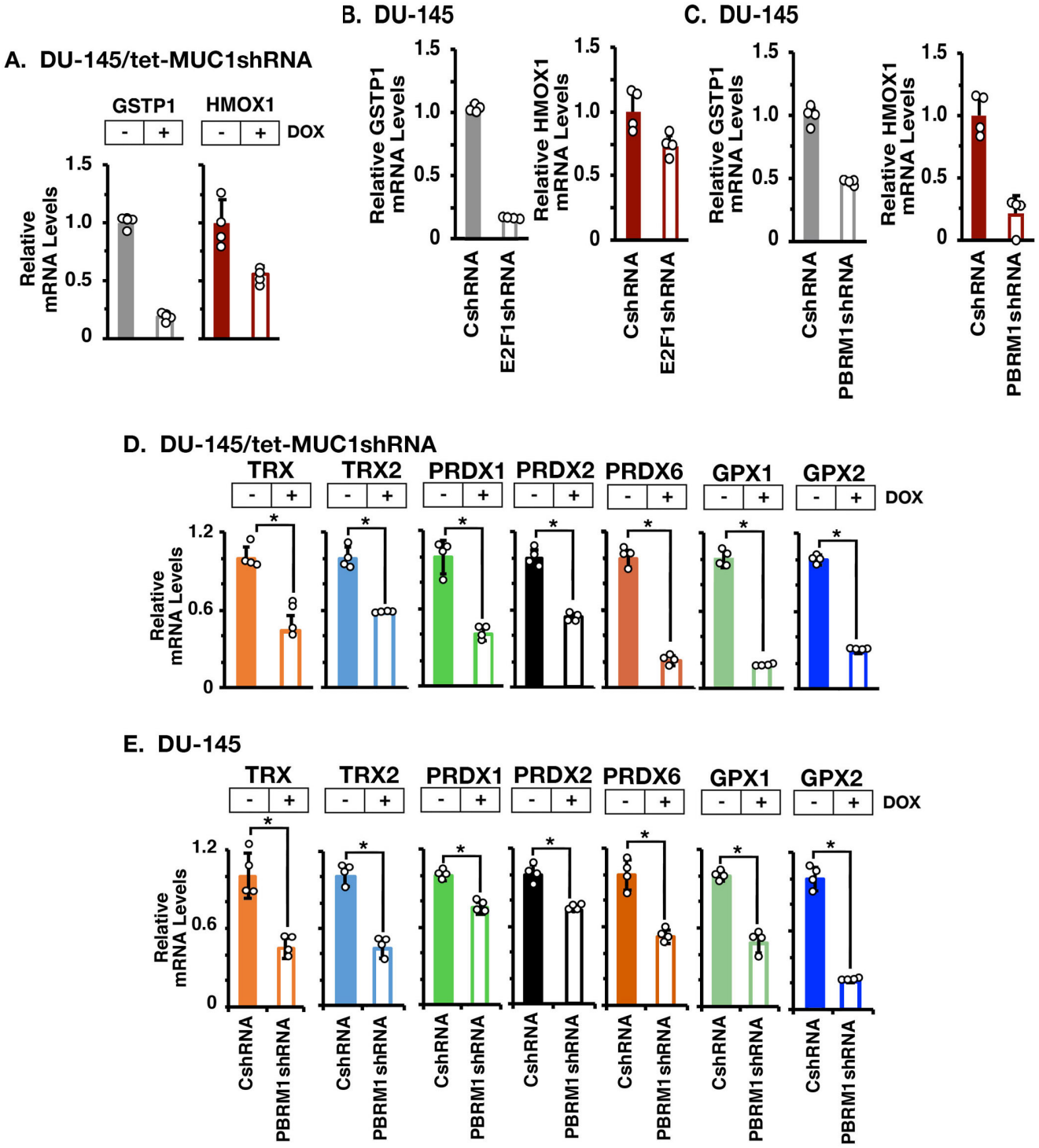


Supplemental Figure S3. Silencing E2F1 downregulates SLC7A11 and G6PD expression. A. Lysates from DU-145/CshRNA and DU-145/MUC1shRNA (left) or DU-145/CsgRNA and DU-145/MUC1sgRNA (right) cells were immunoblotted with antibodies against the indicated proteins. **B.** DU-145/CshRNA and DU-145/E2F1shRNA cells were analyzed for the indicated mRNA levels by qRT-PCR. The results (mean±SD of 3 determinations) are expressed as relative mRNA levels compared to that obtained for CshRNA cells (assigned a value of 1). **C.** Lysates were immunoblotted with antibodies against the indicated proteins. **D.** Lysates from DU-145/CshRNA and DU-145/E2F1shRNA#2 cells were immunoblotted with antibodies against the indicated proteins. **E.** Soluble chromatin from DU-145/tet-MUC1shRNA cells treated with vehicle or DOX for 7 days was precipitated with anti-NRF2 (left), anti-PBRM1 (right) or a control IgG. The DNA samples were amplified by qPCR with primers for the *G6PD*

promoter region. The results (mean±SD of 3 determinations) are expressed as fold enrichment relative to that obtained with the IgG control (assigned a value of 1).

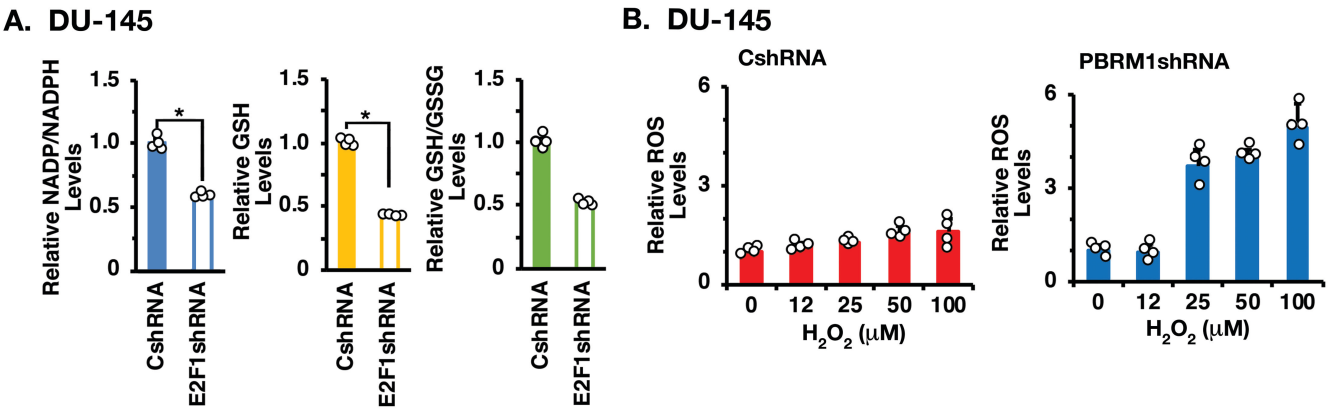

A.
PGD
+1
5' UTR
+1540
Intron-1
Intron-2
Intron-3
+4930
+4938
TGACAGTGC
ChIP
Fold Enrichment Relative to IgG
IgG
NRF2
MUC1-C
PBRM1
ChIP: NRF2 Re-ChIP
B. pPGD
C. DU-145/tet-MUC1-C
DOX
Relative PGD mRNA Levels
kDa
PGD
β-actin
D. DU-145
CshRNA
E2F1shRNA
E. DU-145
PBRM1shRNA
Relative PGD RNA Levels

Supplemental Figure S4. MUC1-C and PBRM1 activate the *PGD* gene. **A.** Schema of the *PGD* promoter region with highlighting of the NRF2 binding site in intron-3. Soluble chromatin from DU-145 cells was precipitated with anti-NRF2, anti-MUC1-C, anti-PBRM1 or a control IgG (left). Soluble chromatin from DU-145 cells was precipitated with anti-NRF2 (ChIP) and then reprecipitated with anti-MUC1-C, anti-PBRM1 or a control IgG (re-ChIP)(right). The DNA samples were amplified by qPCR with primers for the *PGD* promoter region. The results (mean±SD of 3 determinations) are expressed as fold enrichment relative to that obtained with the IgG control (assigned a value of 1). **B.** Chromatin from DU-145/tet-MUC1shRNA cells treated with vehicle or DOX for 7 d was analyzed for ATAC-seq. UCSC genome browser snapshot of ATAC-seq data from the *PGD* gene showing loss of peaks and decrease in chromatin accessibility as a function of MUC1-C silencing. **C-E.** DU-145/tet-MUC1shRNA cells treated with vehicle or DOX for 7 days (**C**), DU-145/CshRNA and DU-145/E2F1shRNA (**D**) or DU-145/CshRNA and DU-145/PBRM1shRNA (**E**) cells were analyzed for PGD mRNA levels by qRT-PCR. The results (mean±SD of 4 determinations) are expressed as relative mRNA levels compared to that obtained for CshRNA cells (assigned a value of 1)(left). Lysates were immunoblotted with antibodies against the indicated proteins (right).



Supplemental Figure S5. MUC1-C, E2F1 and PBRM1 drive NRF2 target antioxidant genes. A. 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 obtained for vehicle-treated cells (assigned a value of 1). **B and C.** DU-145/CshRNA, DU-145/E2F1shRNA (**B**) and DU-145/PBRM1shRNA (**C**) 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 obtained for CshRNA cells (assigned a value of 1). **D.** 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 obtained for vehicle-treated cells (assigned a value of 1). **E.** DU-145/CshRNA and DU-145/PBRM1shRNA 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 obtained for CshRNA cells (assigned a value of 1).



Supplemental Figure S6. Effects of silencing E2F1 and PBRM1 on redox balance. A. DU-145/CshRNA and DU-145/E2F1shRNA cells were analyzed for NADP/NADPH (left), GSH (middle) and GSH/GSSG (right) levels. The results (mean±SD of 4 determinations) are expressed as relative levels compared to that obtained for CshRNA cells (assigned a value of 1). **B.** DU-145/CshRNA (left) and DU-145/PBRM1shRNA (right) cells were incubated with the indicated H_2O_2 concentrations for 1 hour and analyzed for ROS levels. The results (mean±SD of 4 determinations) are expressed as relative ROS levels compared to that obtained for control cells (assigned a value of 1).

Supplemental Tables

Table S1. Primers used for qRT-PCR.

MUC1-C	**FWD**	TACCGATCGTAGCCCCTATG
	REV	CTCACCAGCCCAAACAGG
PBRM1	**FWD**	AAGAAGAAAGAGCTTGCCAG
	REV	TCTCGAGCTTCAAGAACAAC
ARID2	**FWD**	GCAGCCAATTTCCACTCCTGTTG
	REV	GATTGGTGACAGGAGTCCTCTG
BRD7	**FWD**	CAAGCTCTTTAGCCAAACAAGAA
	REV	TCATTCCTGAGTGCAACAGC
E2F1	**FWD**	TATGGTGATCAAAGCCCCTC
	REV	AGATGATGGTGGTGGTGACA
SLC7A11	**FWD**	CCATGAACGGTGGTGTGTT
	REV	GACCCTCTCGAGACGCAAC
G6PD	**FWD**	TGCCTTCCATCAGTCGGATACA
	REV	TGGTGGGGTAGATCTTCTTCTTGG
PGD	**FWD**	GGCTTTGTGGTCTGTGCTTT
	REV	AAATCATCCACAGCTTGCCC
GSTP1	**FWD**	ACCCCAGGGCTCTATGGGAA
	REV	TGAGGGCACAAGAAGCCCCT
HMOX1	**FWD**	CAGCATGCCCCAGGATTTG
	REV	AGCTGGATGTTGAGCAGGA
TRX	**FWD**	CGCGGATCCATGGTGAAGCAGATCG
	REV	CCGCTCGAGTTAGACTAATTCATTA
TRX2	**FWD**	CGCGGATCCATGGCTCAGCGACTTC
	REV	CCGCTCGAGTCAGCCAATCAGCTTC
PRDX1	**FWD**	TTTGGTATCAGACCCGAAGC
	REV	TCCCCATGTTTGTCAGTGAA
PRDX2	**FWD**	GTCCGTGCGTCTAGCCTTT
	REV	TCCCTTTGTAGTCCGACAGC
PRDX6	**FWD**	GGACGTGGCTCCCAACTTT
	REV	CGAGGGTGGGAGAAGAGAATG

GPX1	**FDW**	AAGGTACTACTTATCGAGAATGTG
	REV	GTCAGGCTCGATGTCAATGGTCTG
GPX2	**FWD**	GACACGAGGAAACCGAAGCA
	REV	GGCCCTTCACAACGTCT
GAPDH	**FWD**	CCATGGAGAAGGCTGGGG
	REV	CAAAGTTGTCATGGATGACC

Table S2. Primers used for ChIP-qPCR.

pPBRM1	**FWD**	ACTTTCTCACAGCTGCACTC
	REV	GCGGGAAAGTCTGGGTTAAT
pARID2	**FWD**	GAGCTTCCTTTCCCTTCAGAG
	REV	CCGGTTGTTCCAGGGTTAG
pBRD7	**FWD**	CAAGAAGCACAAGAAGCACAAG
	REV	CACTGGGAAAGAGCGGAAG
pSLC7A11	**FWD**	TTGACTATGCCCTGACACATTAG
	REV	ACAGGAAGCCATCTTCTTTCTC
pG6PD	**FWD**	CCTGGGTTCAAGCGATTCT
	REV	GGTGAAACTCCGTCTCTACTAAC
pPGD	**FWD**	GCGAGACTCCGTCTCAAATAA
	REV	ATCTACCTACAGCACACCAAC
GAPDH	**FWD**	TACTAGCGGTTTTACGGGCG
	REV	TCGAACAGGAGGAGCAGAGAGCGA