

Supplementary Materials

Antimetastatic Properties of Prodigiosin and the BH3-Mimetic Obatoclax (GX15-070) in Melanoma

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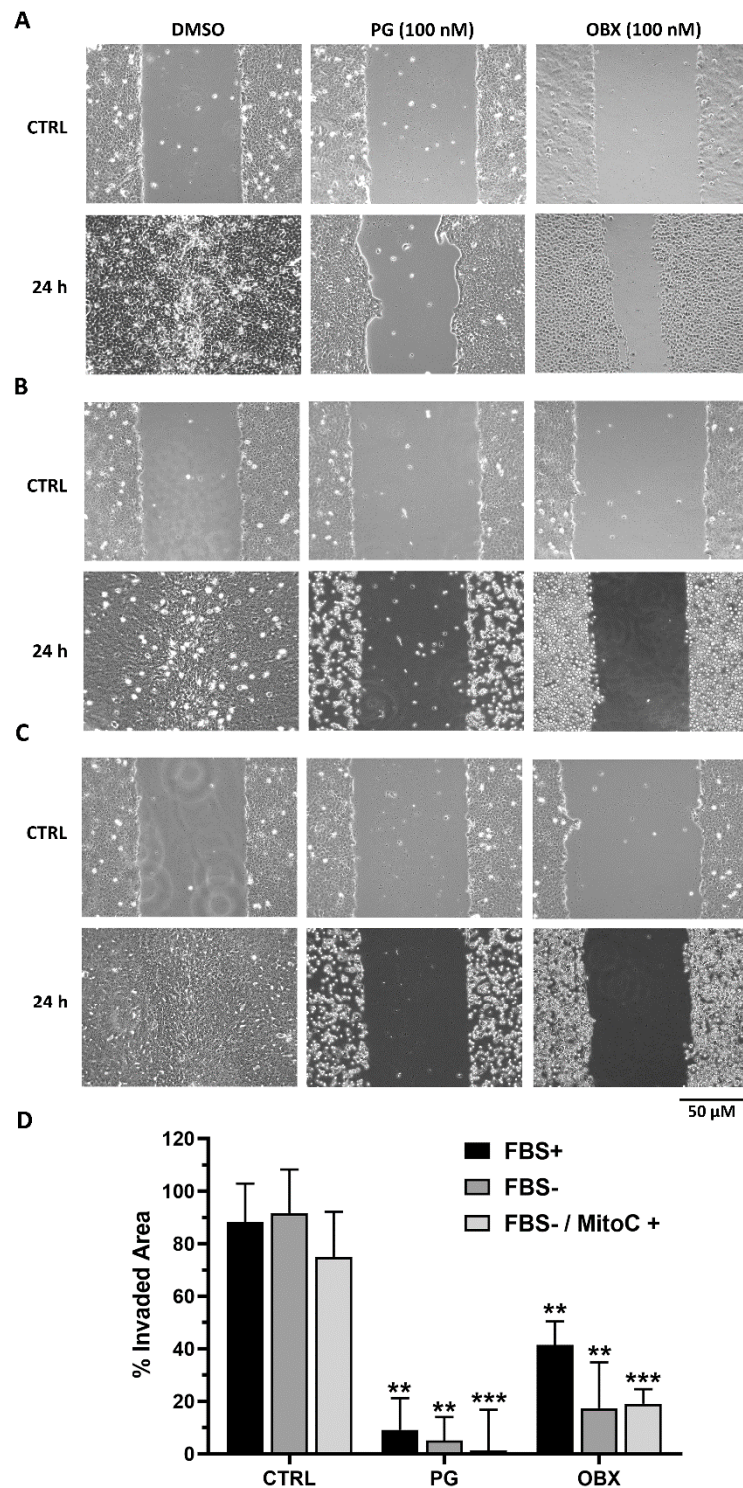
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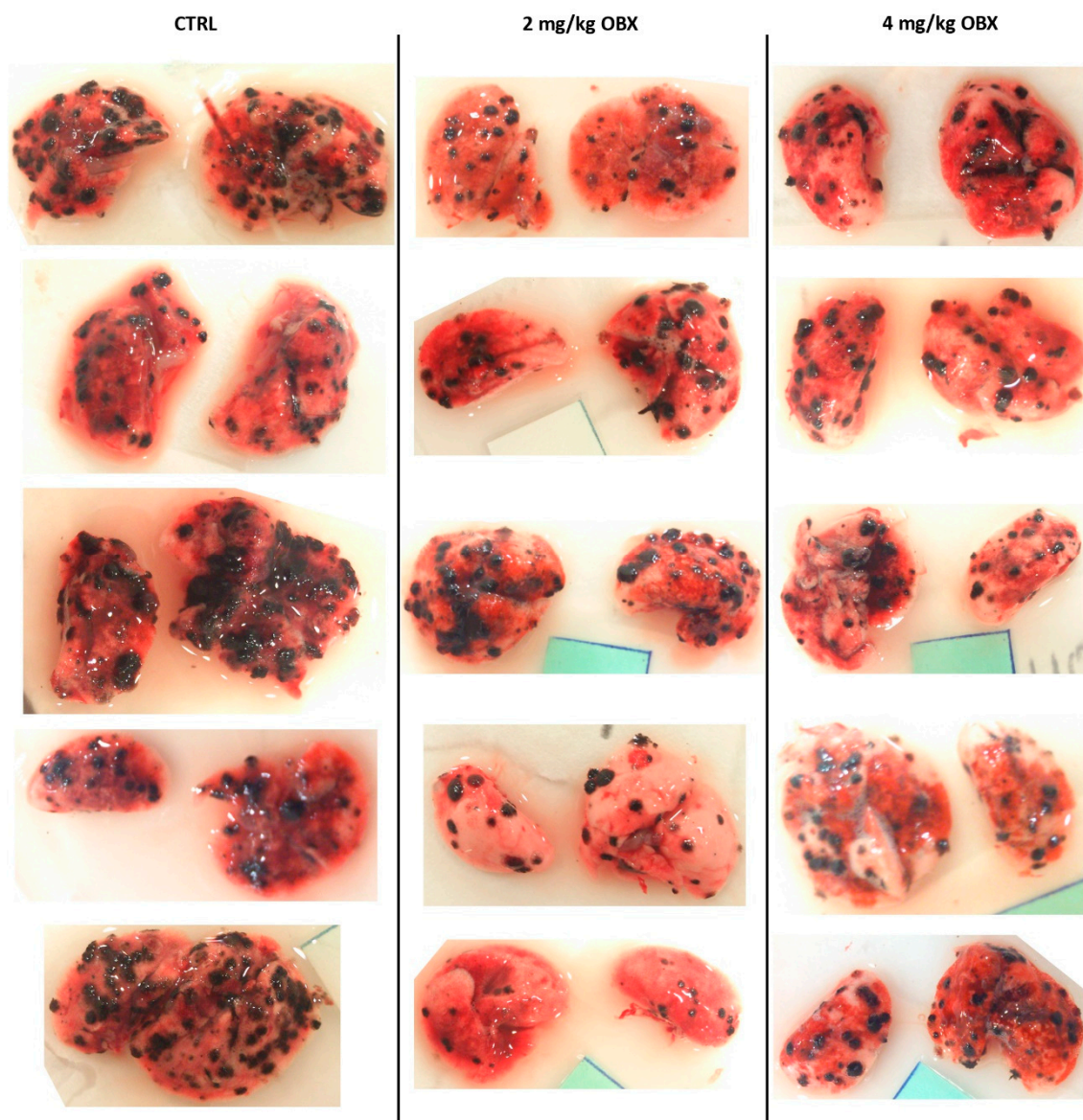
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Supplementary Figures



Supplementary Figure S1. Effects of prodigiosin (PG) and obatoclast (OBX) on HN4 cell migration. The wound healing assay was performed in three conditions: complete media with serum (FBS+) (A), serum deprived (FBS-) (B) and serum deprived plus mitomycin (MitoC) treatment (C). Cells were treated with 100 nM of PG or OBX for 24 h. Representative images of three independent replicates are shown. (D) Quantification of the % invaded area after PG and OBX treatment. Figure shows mean \pm SD. Statistical differences against control (CTRL) are shown as **** $p < 0.0001$ and * $p < 0.05$.



Supplementary Figure S2. In vivo evaluation of obatoclax (OBX) antimetastatic properties. 15 mice were divided into 3 groups. Two groups were treated daily with 2 or 4 mg/kg of OBX for 10 days and the other group was treated with the vehicle (control, CTRL).

Supplementary Tables

Supplementary Table S1. Gene expression profiling of 87 metastasis-related genes after PG treatment. SK-MEL-5 cells were treated with 0.5 μ M PG (IC₂₅ value) during 16 h and gene expression changes were analyzed by TLDA technology (Applied Biosystems) and DataAssist Software. Mean RQ (Relative Quantitation) of treated cells (IC₂₅ SK5 (RQ)) \pm Standard Deviation (SD) are shown in the table below.

DataAssist Software v2.0
Study: SK5 Ct vs IC25
Maximum allowable CT value: 35.0
Include max CT values in calculations: Yes
Exclude outliers among replicates: Yes.
Normalization method: Arithmetic Mean
Selected controls: B2M-Hs99999907_m1
Reference sample: SK5 Ct

Assay	Type	RQ: Relative Quantitation						
		CT SK5 (RQ)	CT SK5 (RQ Min)	CT SK5 (RQ Max)	IC25 SK5 (RQ)	IC25 SK5 (RQ Min)	IC25 SK5 (RQ Max)	SD
ACTB-Hs99999903_m1	Candidate Control	1.00	0.81	1.23	0.13	0.09	0.18	0.07
APC-Hs00181051_m1	Target	1.00	0.81	1.24	0.70	0.60	0.81	0.15
B2M-Hs99999907_m1	Selected Control	1.00	0.79	1.26	1.00	0.83	1.20	0.26
BRMS1-Hs00363036_m1	Target	1.00	0.79	1.27	0.39	0.31	0.49	0.13
CASP8-Hs01018151_m1	Target	1.00	0.85	1.18	0.19	0.13	0.27	0.10
CD44-Hs00153304_m1	Target	1.00	0.82	1.22	1.76	1.64	1.88	0.17
CD82-Hs00174463_m1	Target	1.00	0.78	1.29	0.64	0.48	0.85	0.26
CDH1-Hs00170423_m1	Target	1.00	0.69	1.45	0.52	0.36	0.75	0.28
CHI3L1-Hs00609691_m1	Target	1.00	1.00	1.00	2.55	2.18	2.99	0.57
CTBP1-Hs00179922_m1	Target	1.00	0.87	1.15	0.62	0.46	0.84	0.27
CTNNA1-Hs00426996_m1	Target	1.00	0.83	1.21	0.55	0.43	0.71	0.20
CTSK-Hs00166156_m1	Target	1.00	0.72	1.38	0.56	0.26	1.20	0.67
CXCL12-Hs00171022_m1	Target	1.00	0.89	1.12	0.76	0.59	0.99	0.28
CXCR4-Hs00607978_s1	Target	1.00	0.87	1.15	4.22	3.30	5.39	1.48
DAPK1-Hs00234480_m1	Target	1.00	0.95	1.05	0.42	0.27	0.67	0.28
DCC-Hs00180437_m1	Target	1.00	0.88	1.13	2.17	2.07	2.27	0.14
EPHB2-Hs00362096_m1	Target	1.00	0.87	1.15	0.59	0.41	0.85	0.31
ERBB2-Hs00170433_m1	Target	1.00	0.74	1.36	0.43	0.38	0.49	0.08
FGF2-Hs00266645_m1	Target	1.00	0.77	1.30	0.36	0.29	0.46	0.13
FGFR4-Hs00242558_m1	Target	1.00	0.83	1.21	1.53	1.24	1.89	0.46
FN1-Hs00365058_m1	Target	1.00	0.79	1.27	0.68	0.53	0.86	0.23
FTH1;MT-CO2;OAF-Hs02596865_g1	Target	1.00	0.79	1.26	0.75	0.60	0.93	0.24
FXYD5-Hs00204319_m1	Target	1.00	0.91	1.10	0.71	0.53	0.96	0.30
GAPDH-Hs99999905_m1	Candidate Control	1.00	0.84	1.19	0.23	0.17	0.31	0.10
GNRH1-Hs00171272_m1	Target	1.00	0.37	2.69	0.98	0.61	1.57	0.68
HGF-Hs00300159_m1	Target	1.00	0.69	1.45	2.43	1.56	3.78	1.57
HPSE-Hs00180737_m1	Target	1.00	0.75	1.34	0.94	0.64	1.40	0.54
HRAS-Hs00610483_m1	Target	1.00	0.87	1.15	0.38	0.27	0.54	0.19
HTATIP2-Hs00185131_m1	Target	1.00	0.85	1.18	0.82	0.63	1.07	0.31
IGF1-Hs00153126_m1	Target	1.00	0.87	1.15	0.86	0.63	1.19	0.39
IL18-Hs00155517_m1	Target	1.00	0.70	1.44	0.29	0.24	0.36	0.09
IL1B-Hs00174097_m1	Target	1.00	0.59	1.70	1.31	1.06	1.62	0.40
ITGA7-Hs00174397_m1	Target	1.00	0.58	1.72	1.54	1.27	1.87	0.43
ITGB3-Hs00173978_m1	Target	1.00	0.83	1.21	1.12	0.91	1.38	0.33
KRAS-Hs00270666_m1	Target	1.00	0.72	1.39	0.30	0.18	0.49	0.22
LAMB1-Hs00158620_m1	Target	1.00	0.86	1.17	0.63	0.57	0.69	0.08

MCAM-Hs00174838_m1	Target	1.00	0.79	1.26	0.70	0.54	0.91	0.26
MET-Hs00179845_m1	Target	1.00	0.85	1.17	2.14	1.81	2.54	0.52
MGAT5-Hs00159136_m1	Target	1.00	0.82	1.21	0.59	0.51	0.68	0.12
MITF-Hs00165156_m1	Target	1.00	0.76	1.32	0.32	0.25	0.40	0.11
MMP1-Hs00233958_m1	Target	1.00	0.65	1.54	0.14	0.05	0.38	0.23
MMP10-Hs00233987_m1	Target	1.00	1.00	1.00	2.47	2.24	2.72	0.34
MMP14-Hs01037009_g1	Target	1.00	0.78	1.28	0.86	0.68	1.08	0.28
MMP2-Hs00234422_m1	Target	1.00	0.61	1.63	0.55	0.28	1.09	0.57
MTA1-Hs00183042_m1	Target	1.00	0.86	1.17	0.98	0.75	1.26	0.36
MTSS1-Hs00207341_m1	Target	1.00	0.78	1.28	0.65	0.56	0.76	0.14
MYC-Hs00153408_m1	Target	1.00	0.83	1.20	0.52	0.39	0.69	0.21
NCAM1-Hs00169851_m1	Target	1.00	0.65	1.54	0.84	0.82	0.85	0.02
NF2-Hs00738978_m1	Target	1.00	0.82	1.22	0.29	0.23	0.35	0.08
NME1-Hs02621161_s1	Target	1.00	0.76	1.32	0.27	0.21	0.34	0.09
NR4A3-Hs00235001_m1	Target	1.00	0.82	1.21	0.99	0.84	1.16	0.22
PECAM1-Hs00169777_m1	Target	1.00	0.84	1.19	2.45	1.90	3.16	0.89
PNN-Hs00170192_m1	Target	1.00	0.76	1.31	0.32	0.20	0.54	0.24
PTEN-Hs01920652_s1	Target	1.00	0.72	1.38	0.04	0.02	0.06	0.03
RB1-Hs00153108_m1	Target	1.00	0.69	1.44	0.50	0.41	0.63	0.16
RET-Hs00240887_m1	Target	1.00	0.66	1.52	1.10	0.77	1.57	0.57
RHOA-Hs00357608_m1	Target	1.00	0.78	1.27	0.41	0.33	0.50	0.12
RHOC-Hs00733980_m1	Target	1.00	0.79	1.27	0.84	0.49	1.46	0.69
RORB-Hs00199445_m1	Target	1.00	0.51	1.95	1.15	1.01	1.31	0.21
RYBP-Hs00171928_m1	Target	1.00	0.79	1.26	0.96	0.81	1.14	0.24
SERPINE1-Hs00167155_m1	Target	1.00	0.77	1.29	1.47	0.77	2.81	1.45
SET-Hs00853870_g1	Target	1.00	0.84	1.20	0.41	0.29	0.57	0.20
SMAD2-Hs00183425_m1	Target	1.00	0.81	1.23	0.42	0.33	0.52	0.14
SMAD4-Hs00232068_m1	Target	1.00	0.84	1.19	0.66	0.58	0.76	0.13
SNCG-Hs00268306_m1	Target	1.00	0.45	2.20	1.64	1.13	2.39	0.89
SSTR2-Hs00265624_s1	Target	1.00	0.42	2.39	0.50	0.34	0.73	0.28
SYK-Hs00374292_m1	Target	1.00	0.69	1.44	0.40	0.22	0.72	0.35
TCF20-Hs00390028_m1	Target	1.00	0.77	1.30	0.51	0.41	0.64	0.16
TFRC-Hs99999911_m1	Candidate Control	1.00	0.88	1.14	0.99	0.80	1.23	0.31
TGFB1-Hs99999918_m1	Target	1.00	0.76	1.31	0.57	0.46	0.70	0.17
TGFBR2-Hs00559661_m1	Target	1.00	0.82	1.23	0.33	0.30	0.36	0.04
TIAM1-Hs00180075_m1	Target	1.00	0.86	1.17	0.17	0.12	0.25	0.09
TIMP1-Hs00171558_m1	Target	1.00	0.85	1.17	0.66	0.50	0.88	0.27
TIMP2-Hs00234278_m1	Target	1.00	0.82	1.22	1.06	0.78	1.44	0.47
TIMP3-Hs00165949_m1	Target	1.00	0.81	1.24	0.50	0.39	0.65	0.18
TWIST1-Hs00361186_m1	Target	1.00	0.77	1.30	0.50	0.44	0.58	0.09
UBC-Hs00824723_m1	Candidate Control	1.00	0.83	1.21	0.94	0.73	1.21	0.34
UBE2I-Hs00163336_m1	Target	1.00	0.78	1.28	0.20	0.16	0.27	0.08
VEGF-Hs00900054_m1	Target	1.00	0.81	1.24	2.31	1.90	2.81	0.64
VEGFC-Hs00153458_m1	Target	1.00	0.86	1.16	0.19	0.12	0.29	0.12