Supplementary Figures

Pan-Cancer Genetic Analysis Identifies PARK2 as a Master Regulator of G1/S Cyclins

Yongxing Gong[1], Travis Ian Zack[2,3,4,5,6], Luc G.T. Morris[7], Kan Lin[8], Ellen Hukkelhoven[9], Radhika Raheja[9], I-Li Tan[8], Sevin Turcan[1], Selvaraju Veeriah[1], Shasha Meng[1], Agnes Viale[10], Steven E. Schumacher[2], Perry Palmedo[2,11] , Rameen Beroukhim[2,3,4,5], and Timothy A. Chan[1, 12, 13,14]

[1]Human Oncology and Pathogenesis Program, Memorial Sloan-Kettering Center, New York, NY 10065, USA

[2]The Broad Institute, Cambridge, MA 02142

[3]Dept. of Cancer Biology, [4]Medical Oncology, and [5]Center for Cancer Genome Characterization, Dana-Farber Cancer Institute, Boston, MA 02215

[6]Biophysics Program, Harvard University, Boston, MA 02115

[7]Dept. of Surgery, Memorial Sloan-Kettering Cancer Center, New York, NY 10065, USA.

[8]Weill Cornell College of Medicine, New York, NY 10065.

[9]Molecular Biology Program, [10]Genomics Core, Memorial Sloan-Kettering Cancer Center, New York, NY 10065

[11]Center for Biomedical Informatics, Harvard University, Boston, MA 02115

[12]Dept. of Radiation Oncology, [13]Brain Tumor Center, Memorial Sloan-Kettering Cancer Center, New York, NY 10065

[14]Correspondence should be addressed to: T.A.C. (chant@mskcc.org)

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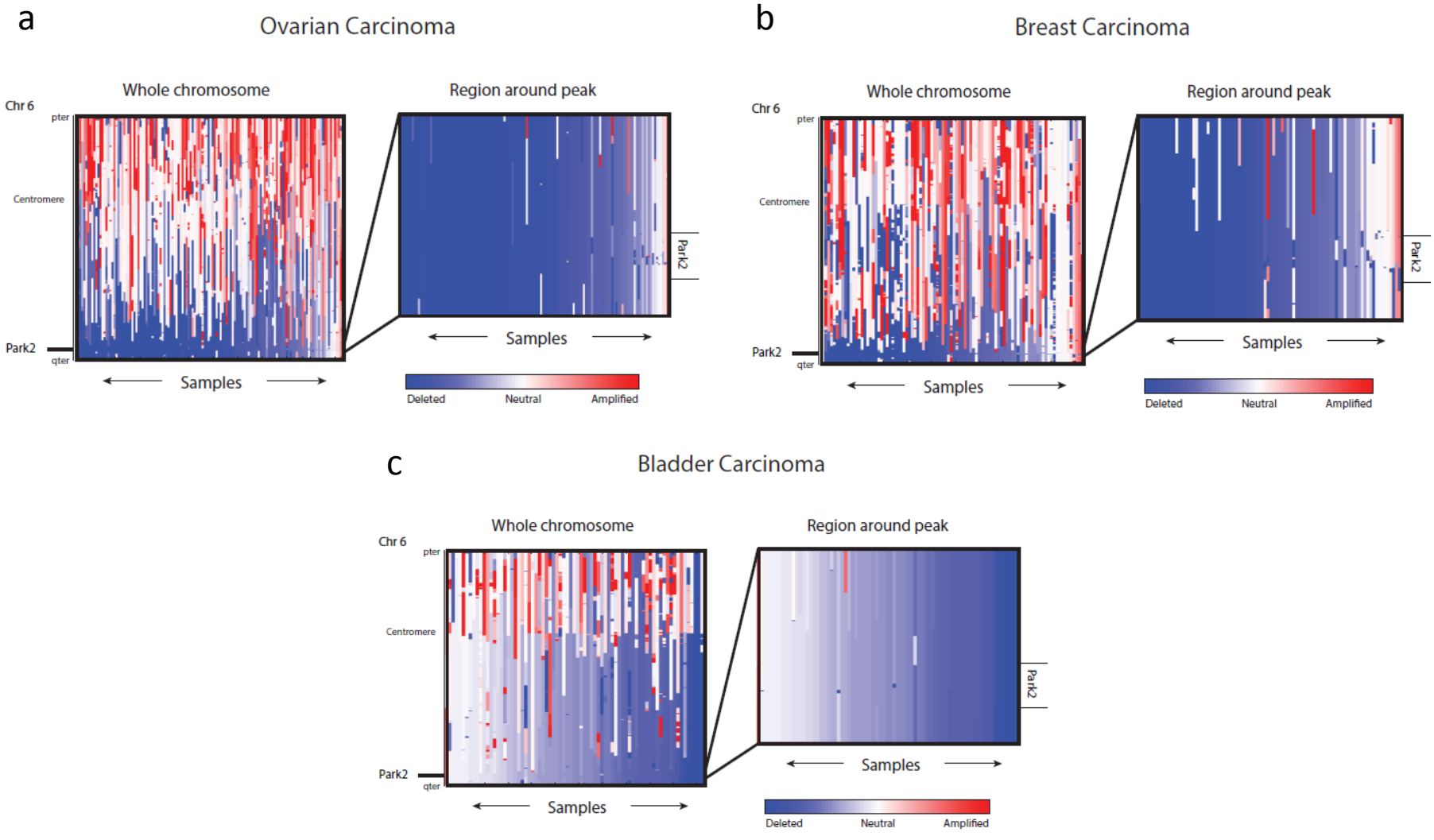

Supplementary Figure 1 Examples of *PARK2* locus profiles from distinct cancer lineages. Copy number data from the TCGA. Representative types of malignancies are shown. SCNA profiles of chromosome 6 (left) and a 5Mb region surrounding *PARK2* (right) are shown in the three lineages that had the highest frequency of *PARK2* deletions: ovarian cancer **(a)**, breast cancer **(b)**, and bladder cancer **(c)**.

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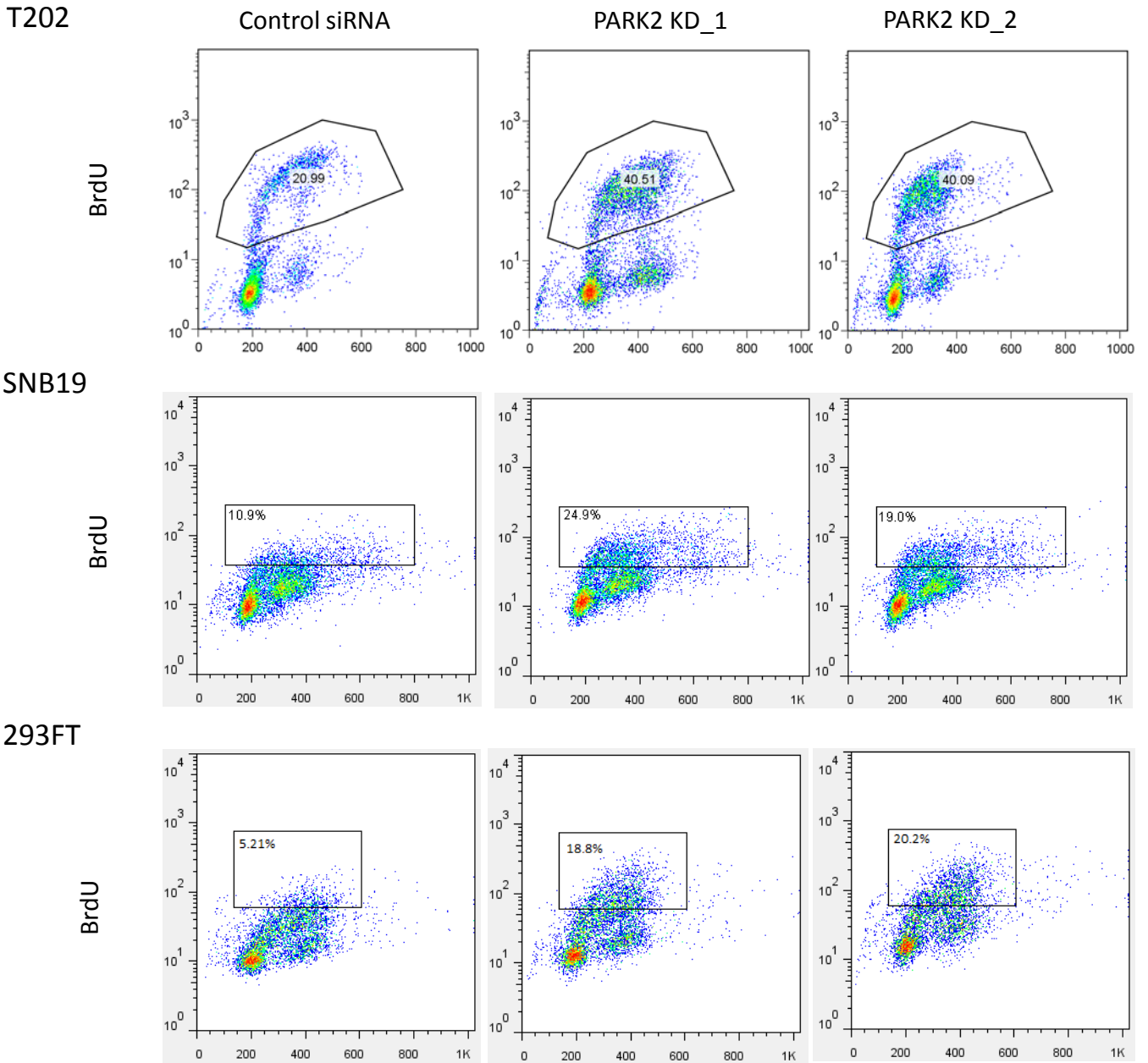

Supplementary Figure 2 PARK2 regulates cell cycle progression. FACS data from three cell lines as indicated. Cells were transfected with scrambled siRNA control or with *PARK2* siRNAs. DNA content and BrdU incorporation were measured after *PARK2* knockdown. Representative results are shown from triplicate experiments.

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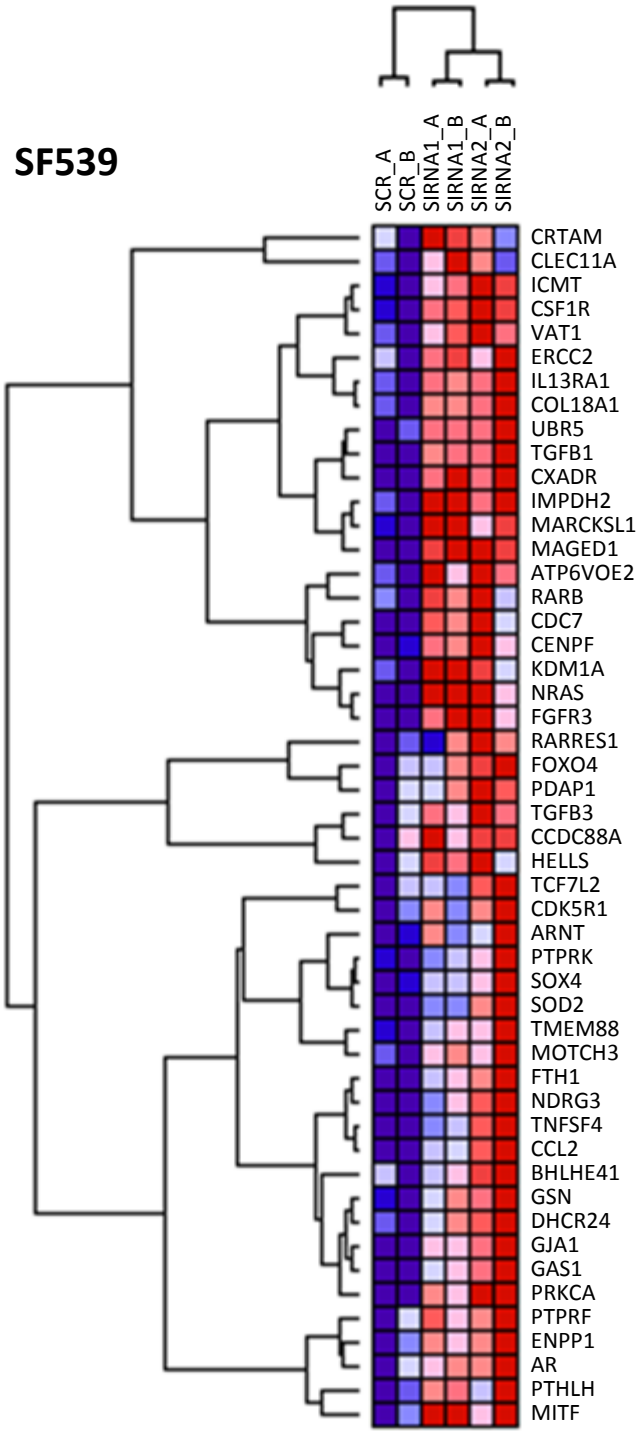
SF539
SCR_A
SCR_B
SIRNA1_A
SIRNA1_B
SIRNA2_A
SIRNA2_B
CRTAM
CLEC11A
ICMT
CSF1R
VAT1
ERCC2
IL13RA1
COL18A1
UBR5
TGFB1
CXADR
IMPDH2
MARCKSL1
MAGED1
ATP6VOE2
RARB
CDC7
CENPF
KDM1A
NRAS
FGFR3
RARRES1
FOXO4
PDAP1
TGFB3
CCDC88A
HELLS
TCF7L2
CDK5R1
ARNT
PTPRK
SOX4
SOD2
TMEM88
MOTCH3
FTH1
NDRG3
TNFSF4
CCL2
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GSN
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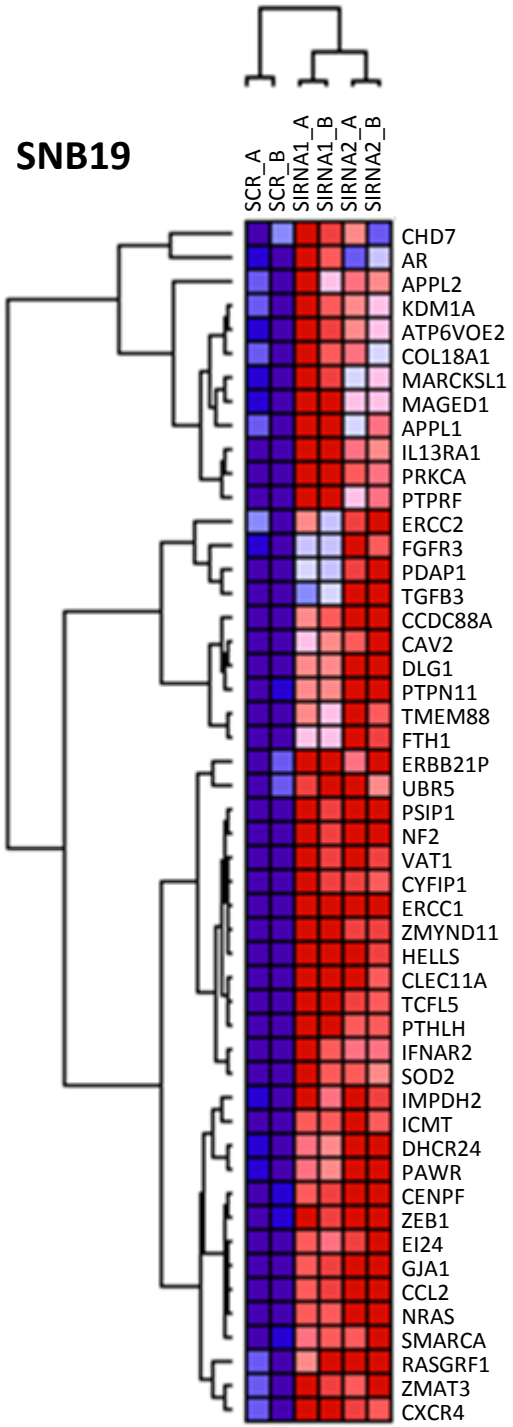
GAS1
PRKCA
PTPRF
ENPP1
AR
PTHLH
MITF


SNB19
SCR_A
SCR_B
SIRNA1_A
SIRNA1_B
SIRNA2_A
SIRNA2_B
CHD7
AR
APPL2
KDM1A
ATP6VOE2
COL18A1
MARCKSL1
MAGED1
APPL1
IL13RA1
PRKCA
PTPRF
ERCC2
FGFR3
PDAP1
TGFB3
CCDC88A
CAV2
DLG1
PTPN11
TMEM88
FTH1
ERBB21P
UBR5
PSIP1
NF2
VAT1
CYFIP1
ERCC1
ZMYND11
HELLS
CLEC11A
TCFL5
PTHLH
IFNAR2
SOD2
IMPDH2
ICMT
DHCR24
PAWR
CENPF
ZEB1
EI24
GJA1
CCL2
NRAS
SMARCA
RASGRF1
ZMAT3
CXCR4

Supplementary Figure 3 *PARK2* knockdown results in alterations in gene expression. The indicated cells were treated with scrambled siRNA or *PARK2* siRNA as shown. The heat map shows the top 50 genes with increased expression.

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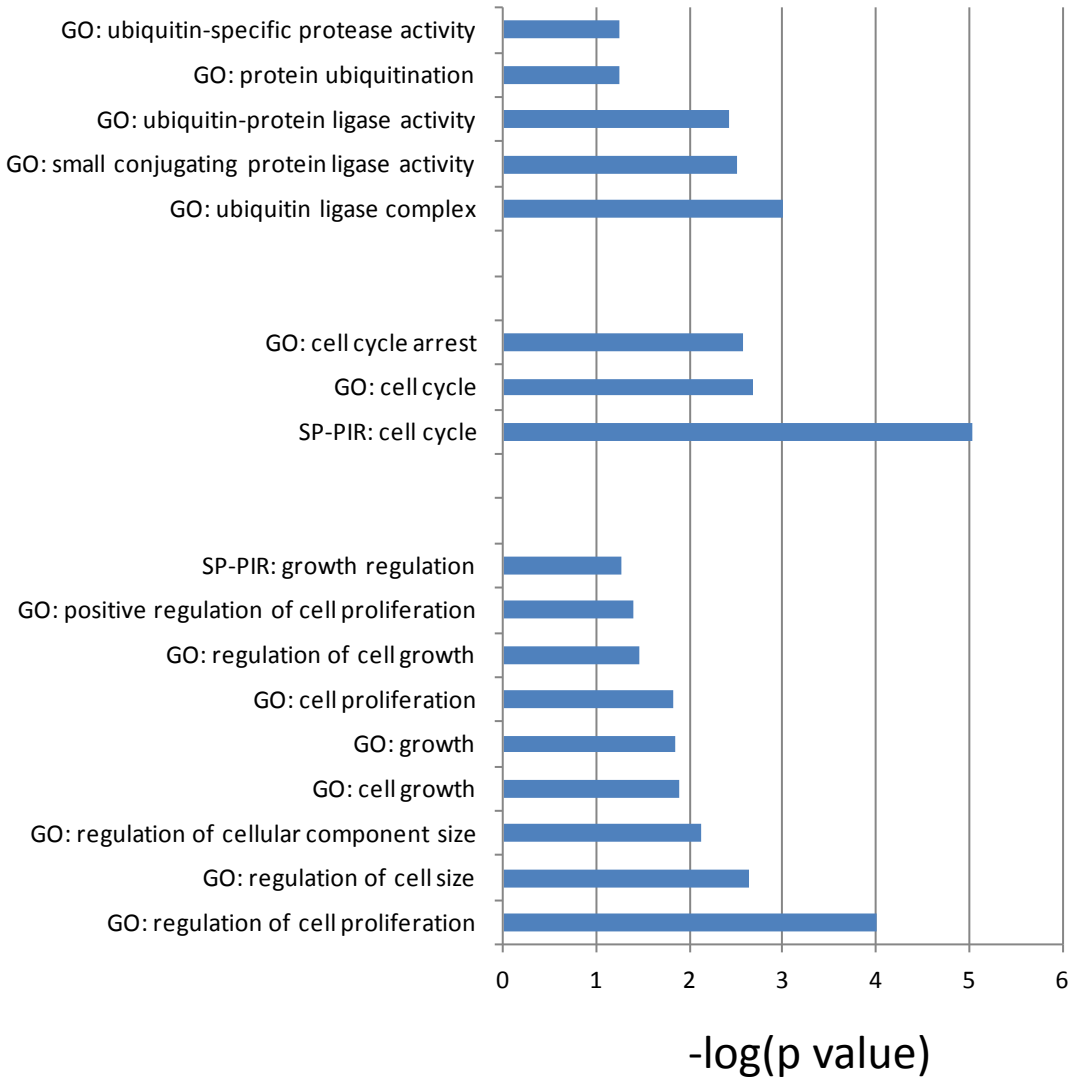

Supplementary Figure 4 Pathway analyses of 2,698 genes differentially expressed across 2 cell lines transfected with *PARK2* siRNAs. Significant enrichment is shown in transcriptional programs governing cell growth, proliferation, protein ubiquitination, and cell cycle control.

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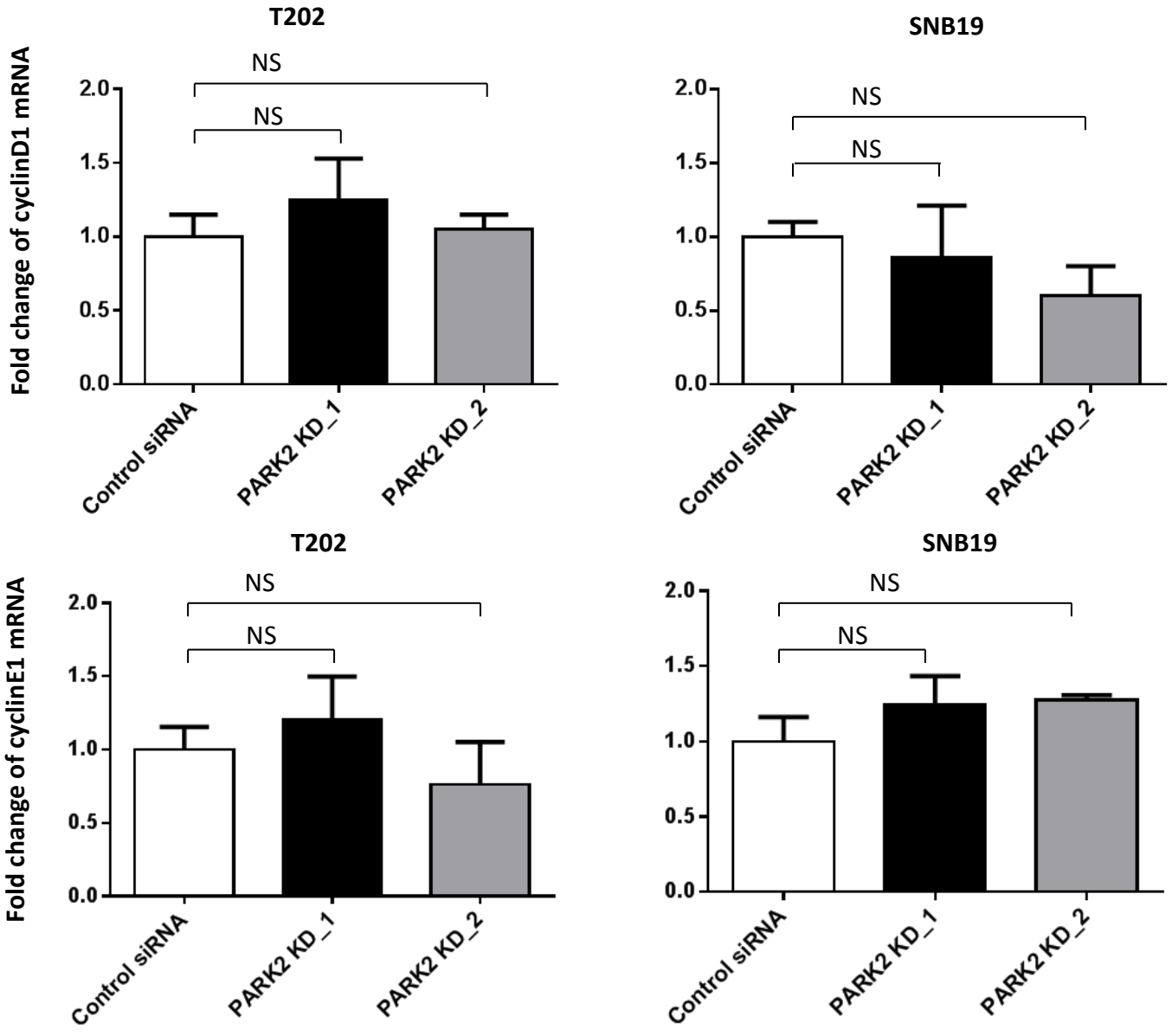
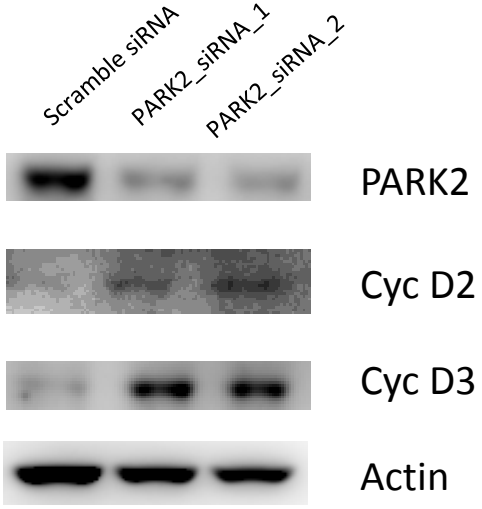


Supplementary Figure 5 Gene expression does not change for cyclins D1 and E1 after *PARK2* knockdown. Two independent *PARK2* siRNAs were used. Experiments were repeated five times. Error bars, 1 standard deviation. NS, not significant.



Supplementary Figure 6 *PARK2* knockdown causes accumulation of cyclin D2 and D3 protein. T202 cells were transfected with scrambled siRNA control or with *PARK2* siRNAs and protein blots were performed with the antibodies specific for each cyclin type as indicated. Representative results are shown from triplicate experiments.

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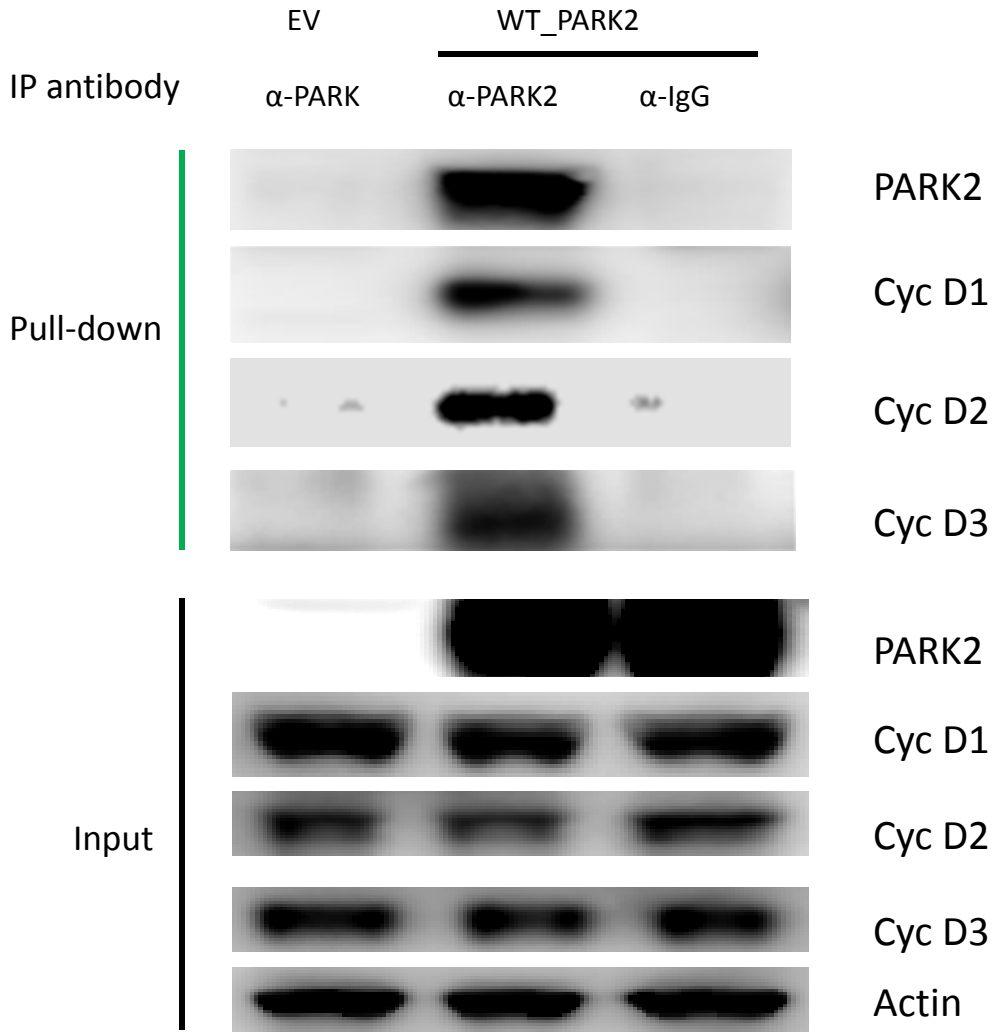

Supplementary Figure 7 Immunoprecipitation assays show binding of wild-type PARK2 to endogenous cyclins D2 and D3. HEK 293T cells were transfected with either vector only (pcDNA3.1) or with vector encoding wild-type PARK2. Assays were performed as described in Online Methods.

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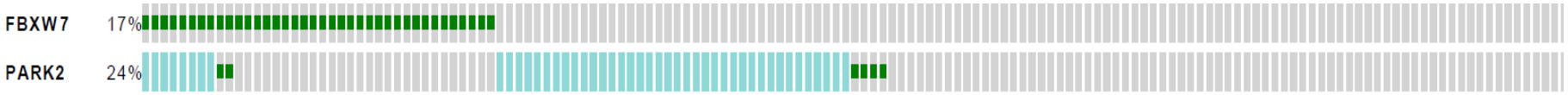

Supplementary Figure 8 Patterns of genetic alteration of *FBXW7* and *PARK2* in colorectal cancer. In colorectal cancer, FBXW7 is primarily mutated while PARK2 is deleted and mutated. Shown are data from The Cancer Genome Atlas Project (http://www.cbioportal.org/public-portal/). Point mutations are noted in green and deletions are noted in blue. Percentages denote total numbers of tumors with alterations in the gene. *p*, not significant.

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Supplementary Table 1. Pan-cancer regions of significant SCNA and PARK2 anticorrelates

A) Amplification

Peak Name	Overall Gistic Rank	Genomic location	Peak region	GISTIC q-value	Gene count	Target(s)	Frequently mutated genes[B]
CCND1	1	11q13.3	chr11:69464719-69502928	2.05E-278	2	*CCND1* [K]	*CCND1* = 6.6e-08
EGFR	2	7p11.2	chr7:55075808-55093954	2.30E-240	1	*EGFR* [K]	*EGFR* = 2.2e-15
MYC	3	8q24.21	chr8:128739772-128762863	6.50E-180	1	*MYC* [K]	
TERC	4	3q26.2	chr3:169389459-169490555	5.40E-117	2	*TERC* [P]	
ERBB2	5	17q12	chr17:37848534-37877201	1.59E-107	1	*ERBB2* [K]	*ERBB2* = 1.3e-06
CCNE1	6	19q12	chr19:30306758-30316875	4.77E-90	1	*CCNE1* [K]	
MCL1	7	1q21.3	chr1:150496857-150678056	1.25E-80	6	*MCL1* [K]	
MDM2	8	12q15	chr12:69183279-69260755	2.59E-62	2	*MDM2* [K]	
INTS4	9	11q14.1	chr11:77610143-77641464	1.01E-54	1	*INTS4*	
CDK4	11	12q14.1	chr12:58135797-58156509	5.14E-41	5	*CDK4* [K]	*CDK4* = 0.0048
SOX2	13	3q26.33	chr3:181151312-181928394	1.21E-38	2	*SOX2* [K]	
PDGFRA	14	4q12	chr4:54924794-55218386	1.08E-37	3	*PDGFRA* [K]	
BDH1	15	3q29	chr3:197212101-197335320	1.21E-31	1	*BDH1* [M]	
MDM4	17	1q32.1	chr1:204367383-204548517	1.98E-29	3	*MDM4* [K]	
TERT	18	5p15.33	chr5:1287704-1300024	9.34E-27	1	*TERT* [K]	
KDM5A	19	12p13.33[T]	chr12:1-980639	1.59E-25	11	*KDM5A* [E]	
MYCL1	20	1p34.2	chr1:40317971-40417342	3.99E-25	2	*MYCL1* [K]	
IGF1R	21	15q26.3	chr15:98667475-100292401	8.62E-25	9	*IGF1R* [K]	
BCL2L1	27	20q11.21	chr20:30179028-30320705	2.85E-15	4	*BCL2L1* [K]	
BRD4	32	19p13.12	chr19:15310246-15428182	5.04E-10	3	*NOTCH3* [P], *BRD4* [P,E]	
KRAS	33	12p12.1	chr12:24880663-25722878	9.47E-10	7	*KRAS* [K]	*KRAS* = 1.5e-14
NKX2-1	34	14q13.2	chr14:35587755-37523513	1.33E-09	14	*NKX2-1* [K]	*NFKBIA* =0.0098, *RALGAPA1* =0.027
ZNF217	36	20q13.2	chr20:52148496-52442225	5.83E-08	1	*ZNF217* [K]	*ZNF217* = 0.0082
AKT1	48	14q32.33	chr14:105182581-105333748	0.00028451	7	*AKT1* [K]	*AKT1* = 1.1e-14
CDK6	49	7q21.2	chr7:92196092-92530348	0.00069831	3	*CDK6* [K]	
6p21.1	50	6p21.1	chr6:41519930-44297771	0.0010459	70		
E2F3	56	6p22.3	chr6:19610794-22191922	0.0033658	7	*E2F3* [K]	
NEDD9	63	6p24.2	chr6:11180426-11620845	0.082606	2	*NEDD9* [K]	
PAX8	69	2q13	chr2:113990138-114122826	0.19717	2	*PAX8* [K]	

B) Deletion

Blue indicates large gene

Peak Name	Overall Gistic Rank	Genomic location	Peak region	GISTIC q-value	Gene count	Target(s)	Frequently mutated genes[B]
CDKN2A	1	9p21.3	chr9:21865498-22448737	0	4	*CDKN2A*[K]	*CDKN2A* = 4.4e-15
STK11	2	19p13.3	chr19:1103715-1272039	1.46E-238	7	*STK11*[K]	*STK11* = 2.5e-13
PDE4D	3	5q11.2	chr5:58260298-59787985	2.02E-143	3	*PDE4D*[L]	
PARK2	4	6q26	chr6:161693099-163153207	5.85E-137	1	*PARK2*[L,K]	
LRP1B	5	2q22.1	chr2:139655617-143637838	4.25E-107	1	*LRP1B*[L]	
CSMD1	6	8p23.2	chr8:2079140-6262191	2.39E-96	1	*CSMD1*[L]	
ARID1A	8	1p36.11	chr1:26900639-27155421	5.74E-87	2	*ARID1A*[K]	*ARID1A* = 1.5e-14
PTEN	9	10q23.31	chr10:89615138-90034038	1.12E-79	2	*PTEN*[K]	*PTEN* = 2.2e-15
WWOX	10	16q23.1	chr16:78129058-79627770	8.14E-76	1	*WWOX*[L]	*WWOX* = 0.092
RB1	11	13q14.2	chr13:48833767-49064807	3.88E-75	2	*RB1*[K]	*RB1* = 1.7e-13
FHIT	17	3p14.2	chr3:59034763-61547330	3.01E-55	1	*FHIT*[L]	
RBFOX1	18	16p13.3	chr16:5144019-7771745	1.00E-45	1	*RBFOX1*[L]	
PTPRD	19	9p24.1	chr9:8310705-12693402	3.24E-38	1	*PTPRD*[L]	
FAT1	21	4q35.2	chr4:187475875-188227950	6.81E-36	1	*FAT1*[K]	*FAT1* = 2.4e-15
NF1	26	17q11.2	chr17:29326736-29722618	6.59E-23	5	*NF1*[K]	*NF1* = 3.3e-13
MACROD2	27	20p12.1	chr20:14302876-16036135	9.00E-19	3	*MACROD2*[L]	
5q15	33	5q15	chr5:73236070-114508587	8.15E-13	156	*APC*[K], *CHD1*[E]	*APC* =2.6e-13, *RASA1* =0.0029
MLL3	34	7q36.1	chr7:151817415-152136074	9.26E-13	1	*MLL3*[K,E]	*MLL3* = 1.1e-05
IKZF2	41	2q34	chr2:211542637-214143899	3.24E-09	4	*IKZF2*[K], ERBB4[L]	*ERBB4* = 0.00058
CNTN4	42	3p26.3[T]	chr3:1-3100786	6.44E-09	3	*CNTN4*[L]	
3p12.2	43	3p12.2	chr3:75363575-86988125	1.22E-07	12	*ROBO1*[L], *CADM2*[L]	
RAD51B	44	14q24.1	chr14:68275375-69288431	1.38E-07	2	*RAD51B*[L]	*ZFP36L1* = 0.0016
11q23.1	45	11q23.1	chr11:105849158-117024891	5.31E-07	84	*ATM*[K]	*ATM* =1.4e-06, *POU2AF1* =0.082
IMMP2L	46	7q31.1	chr7:109599468-111366370	5.74E-07	2	*IMMP2L*[L]	
NEGR1	47	1p31.1	chr1:71699756-74522473	7.25E-07	2	*NEGR1*[L]	
BRCA1	48	17q21.31	chr17:41178765-41336147	7.25E-07	2	*BRCA1*[K]	*BRCA1* = 3.5e-08
9q34.3	49	9q34.3	chr9:135441810-139646221	8.73E-06	94	*NOTCH1*[K], *BRD3*[E], *GTF3C4*[E]	*NOTCH1* =1e-08, *RXRA* =2.1e-05, *COL5A1* =0.0022, *TSC1* =0.012

ANKS1B	50	12q23.1	chr12:99124001-100431272	8.73E-06	2	*ANKS1B* [L]	
DMD	51	Xp21.2	chrX:30865118-34644819	5.15E-05	4	*DMD* [L]	
PRKG1	53	10q11.23	chr10:52644085-54061437	9.79E-05	3	*PRKG1* [L]	
AGBL4	55	1p33	chr1:48935280-50514967	0.000219	2	*AGBL4* [L]	
CDKN1B	56	12p13.1	chr12:12710990-12966966	0.00035777	5	*CDKN1B* [K]	*CDKN1B* = 2.2e-06
SMAD4	63	18q21.2	chr18:48472083-48920689	0.036866	3	*SMAD4* [K]	*SMAD4* = 6.6e-15

Notes

[B]GENE = p-value from [Lawrence et al. *upublished data*] corrected to FDR within peak

[K]Known frequently amplified oncogene or deleted TSG

[P]Putative cancer gene

[E]Epigenetic regulator

[M]Mitochondria-associated gene

**Immediately adjacent to peak region

[T]Adjacent to telomere or centromere of acrocentric chromosome

Supplementary Table 2. Significance of PARK2 anticorrelates

A) Amplification

p-value	gene1	gene2	q-value	
0.012143	PAX8	PARK2		0.181759
0.004796	CCND1	PARK2		0.113663
0.016531	CDK4	PARK2		0.215656
0.0001	CCNE1	PARK2		0.009673
0.000204	BCL2L1	PARK2		0.016122

B) Deletion

p-value	gene1	gene2	FDR p-value	
0.004184	PARK2	BRCA1		0.108167

Note: q-value of <0.25 is significant

Supplementary Table 5. Quantitative RT-PCR Primer and siRNA target sequences

Name	**Oligonucleotide sequence (5' -> 3')**
siRNAs	
siPARK2_1	AGTGCCGTATTTGAAGCCTCAGGAACAAC
siPARK2_2	GTACGCTTCTTTACATTCCCGGCAGAAGG
siFBX4_1	GCCGGTACAGTGTGATTCCACAGATTCAA
siFBX4_2	GGCATTGAGTGGATTCTTGAAGAAGTGGA
siFBXW7_1	ACAGGACAGTGTTTACAAATT
siFBXW7_2	GTGAAGTTGTTGGAGTAGATT
qPCR primers	
cyclin D1_F	CTGGCCATGAACTACCTGGA
cyclin D1_R	GTCACACTTGATCACTCTGG
cyclin E1_F	GTTATAAGGGAGACGGGGAG
cyclin E1_R	TGCTCTGCTTCTTACCGCTC
GAPDH_F	AATGAAGGGGTCATTGATGG
GAPDH_R	AAGGTGAAGGTCGGAGTCAA