

The Maternal Embryonic Leucine Zipper Kinase (MELK) is Upregulated in High-Grade Prostate Cancer

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Supplementary methods:

Tissue specimens and clinical data

Immediately after surgery, the prostate specimens were rapidly processed for histopathological analysis and stored in Tissue-Tek OCT (Sakura, Alphen, Netherlands), embedded cryo-blocks at -80°C or in paraffin-embedded formalin-fixed tissue blocks (FFPE) at room temperature, respectively. RNA samples were isolated from paired tumor and non-malignant (benign) tissue samples from a radical prostatectomy cohort with Gleason scores (GS) between 6 and 10 (Table 1).

All tumor and control areas were assigned by an experienced uropathologist (G.S.) who used a HE stained section for selecting appropriate tissue regions. Non-malignant, benign areas were chosen well separated from tumor areas. Several 10 µm cryo- or FFPE sections were prepared and the assigned tissue was collected by macrodissection using the HE stain as a template. Detailed clinical data are given in Supplementary Table 1.

To this day the Gleason score reflecting the histomorphological pattern and degree of dedifferentiation of prostate tumors is the gold standard of prostate cancer categorization and the best available single parameter for estimation of the risk for progression and worse prognosis [1, 2]. The Gleason system categorizes prostate tumors according to the histomorphological pattern such as loss of a differentiated glandular structure into Gleason patterns (GP) 1 (fully differentiated) to 5 (fully dedifferentiated). The Gleason Score is formed by the sum of the most (primary) and the second most (secondary) predominant Gleason patterns of a tumor and ranges from 2 to 10 [3, 4].

The criteria on PSA-density, biopsy Gleason Score and tumor extension defined by Epstein were used to assign tumor cases into clinically significant and insignificant groups [5-7]. Patients with an ERG gene rearrangement were assessed using a FISH break-apart assay [8]. Alternative approaches for tumor classification risk groups are based on genetic alterations, above all the TMPRSS2-ERG gene rearrangement that is found in about 50% of all prostate tumors [9, 10].

Western blotting

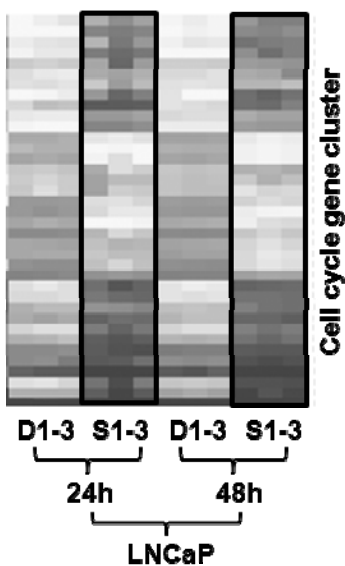
The following primary antibodies were used: Anti-MELK (HPA017214, Sigma), Anti-UBE2C (ab56861, Abcam, Cambridge, UK), Anti-FOXM1 (ab55006, Abcam), Anti-CDC20 (4823, Cell Signaling, Danvers, MA, USA) Anti-AURKA (HPA002636, Sigma), Anti-AR (sc-816, Santa Cruz), Anti-pAKT (9271, Cell Signaling), Anti-Tubulin, alpha (2144, Cell Signaling, Loading control).

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Supplementary Figure 1: Microarray analysis of Siomycin A treated LNCaP indicates specific pattern of responsive genes. Siomycin A treated cells (biological triplicates, S1-3) are compared to DMSO controls (C1-3) for two time points. Here, expression of 33 cell cycle genes coexpressed with MELK and upregulated in aggressive prostate cancer is visualized by hierarchical clustering. In LNCaP cells the majority of these genes (25 out of 33) were downregulated after 24h and 48h Siomycin A treatment.



Supplementary Tables:

Supplementary data is given for detailed lists of differentially expressed genes, ontology statistics and assay information. For Illumina data universal probe ID (Ref ID) is given in all gene lists including statistical analysis outcome (LIMMA software, p-value, linear fold change). Gene ontology statistics was done using GOstat software for down- and upregulated genes, respectively. Results were only documented if significant biological processes were associated with these genes.

Val_32	tumor and benign	low	7	3	4	2c	x	x	0	N/A	N/A	50	9.9	10	36	7	3	4	18	60	0.55	N/A	N/A	N/A
Val_33	tumor and benign	low	5	3	2	2c	x	x	0	N/A	N/A	60	3.9	16	25	6	3	3	29	20	0.13448276	N/A	N/A	N/A
Val_34	tumor and benign	low	5	3	2	2c	x	x	0	N/A	N/A	67	9.03	15	38	6	3	3	40	26.6666667	0.22575	N/A	N/A	N/A
Val_35	tumor and benign	low	5	3	2	2c	x	x	0	N/A	N/A	55	4	10	45	6	3	3	46	6.6666667	0.08695652	N/A	N/A	N/A
Val_36	tumor and benign	low	6	3	3	2c	x	x	0	N/A	N/A	61	3.5	15	30	6	3	3	30	40	0.11666667	N/A	N/A	N/A
Val_37	tumor and benign	low	6	3	3	2a	x	x	0	N/A	N/A	56	2.8	15	34	6	3	3	28	6.6666667	0.1	N/A	N/A	N/A
Val_38	tumor and benign	low	6	3	3	2c	x	x	1	N/A	N/A	61	10.78	7	45	6	3	3	50	6.6666667	0.2156	N/A	N/A	N/A
Val_39	tumor and benign	low	7	3	4	2c	0	x	x	N/A	N/A	59	9	10	42	6	3	3	27	20	0.33333333	N/A	N/A	N/A
Val_40	tumor and benign	low	6	3	3	2c	0	x	0	N/A	N/A	54	7.8	10	32	6	3	3	17	50	0.45882353	N/A	N/A	N/A
Val_41	tumor and benign	low	5	3	2	2c	0	x	0	N/A	N/A	58	7.9	9	40	5	2	3	25	10	0.316	N/A	N/A	N/A
Val_42	tumor and benign	low	6	3	3	2a	0	x	0	N/A	N/A	65	2.1	18	55	6	3	3	42	20	0.05	N/A	N/A	N/A
Val_43	tumor and benign	low	5	2	3	2c	0	x	0	N/A	N/A	57	2.6	12		7	3	4	20	20	0.13	N/A	N/A	N/A
Val_44	tumor and benign	low	7	3	4	2c	x	x	1	N/A	N/A	63	3.77	17	30	7	3	4	42	26.6666667	0.0897619	N/A	N/A	N/A
Val_45	tumor and benign	low	6	3	3	2c	x	x	0	N/A	N/A	64	3.64	14	48	6	3	3	50	6.6666667	0.0728	N/A	N/A	N/A
Val_46	tumor and benign	low	6	3	3	2c	x	x	0	N/A	N/A	64	4.27	15	30	6	3	3	35	6.6666667	0.122	N/A	N/A	N/A
Val_47	tumor and benign	low	6	3	3	2c	0	x	0	N/A	N/A	49	5.3	6	20	5	2	3	17	30	0.31176471	N/A	N/A	N/A
Val_48	tumor and benign	low	5	2	3	2c	0	x	0	N/A	N/A	62	5.4	12	47	5	2	3	39	2.85714286	0.13846154	N/A	N/A	N/A
Val_49	tumor and benign	low	6	3	3	2c	0	x	0	N/A	N/A	52	3.6	14	44	6	3	3	28	50	0.12857143	N/A	N/A	N/A
Val_50	tumor and benign	low	5	3	2	2c	x	x	0	N/A	N/A	57	4.88	12	30	6	3	3	40	6.6666667	0.122	N/A	N/A	N/A
Val_51	tumor and benign	low	5	3	2	2c	0	x	0	N/A	N/A	54	4.2	22	37	6	3	3	30	10	0.14	N/A	N/A	N/A
Val_52	tumor and benign	high	9	4	5	3a	0	x	0	N/A	N/A	67	6.91	6	27	7	3	4	20	46.6666667	0.3455	N/A	N/A	N/A
Val_53	tumor and benign	low	6	3	3	2a	0	x	0	N/A	N/A	48	2	10		6	3	3	22	20	0.09090909	N/A	N/A	N/A
Val_54	tumor and benign	low	7	3	4	3a	0	x	0	N/A	N/A	59	2.1	16	20	6	3	3	20	18.1818182	0.105	N/A	N/A	N/A
Val_55	tumor and benign	low	6	3	3	3a	0	x	0	N/A	N/A	56	3.1	5	32	6	3	3	17	30	0.18235294	N/A	N/A	N/A
Val_56	tumor and benign	low	6	3	3	2c	0	x	1	N/A	N/A	55	8.3	6		4	2	2	29	10	0.2862069	N/A	N/A	N/A
Val_57	tumor and benign	high	9	4	5	3a	x	x	0	N/A	N/A	65	2.65	17	30	6	3	3	39	6.6666667	0.06794872	N/A	N/A	N/A
Val_58	tumor and benign	low	5	2	3	2b	0	x	0	N/A	N/A	57	8.2	11		6	3	3	31	30	0.26451613	N/A	N/A	N/A
Val_59	tumor and benign	low	5	2	3	2b	0	x	0	N/A	N/A	50	4.5	13	48	6	3	3	24	33.3333333	0.1875	N/A	N/A	N/A
Val_60	tumor and benign	low	6	3	3	2b	x	x	0	N/A	N/A	63	6.28	6	32	6	3	3	28	6.6666667	0.22428571	N/A	N/A	N/A
Val_61	tumor and benign	low	5	3	2	2c	x	x	0	N/A	N/A	68	7.27	40	70	6	3	3	53	26.6666667	0.13716981	N/A	N/A	N/A
Val_62	tumor and benign	high	7	4	3	3a	0	x	0	N/A	N/A	67	6.1	20	30	7	3	4	25	46.6666667	0.244	N/A	N/A	N/A
Val_63	tumor and benign	low	7	3	4	2c	x	x	0	N/A	N/A	60	2.2	10	30	6	3	3	28	6.6666667	0.07857143	N/A	N/A	N/A
Val_64	tumor and benign	low	7	3	4	3a	0	x	1	N/A	N/A	60	6.45	10	31	6	3	3	25	66.6666667	0.258	N/A	N/A	N/A
Val_65	tumor and benign	high	8	4	4	3b	x	x	1	N/A	N/A	68	20.75	10	60	6	3	3	50	13.3333333	0.415	N/A	N/A	N/A
Val_66	tumor and benign	high	9	5	4	3b	0	x	0	N/A	N/A	67	5.9	15	61	7	4	3	28	40	0.21071429	N/A	N/A	N/A
Val_67	tumor and benign	low	5	2	3	2b	0	x	0	N/A	N/A	67	8.5	15	62	4	2	2	47	14.2857143	0.18085106	N/A	N/A	N/A
Val_68	tumor and benign	low	6	3	3	2b	x	x	0	N/A	N/A	69	7.3	13		6	3	3	26	11.11111111	0.28076923	N/A	N/A	N/A
Val_69	tumor and benign	low	7	3	4	2a	0	x	0	N/A	N/A	53	2.2	11	40	6	3	3	25	10	0.088	N/A	N/A	N/A
Val_70	tumor and benign	low	5	2	3	2b	0	x	0	N/A	N/A	57	6.2	13		7	3	4	40	30	0.155	N/A	N/A	N/A
Val_71	tumor and benign	high	9	4	5	3b	x	x	1	N/A	N/A	72	18.09	9	53	8	3	5	40	26.6666667	0.45225	N/A	N/A	N/A
Val_72	tumor and benign	high	8	3	5	2b	0	x	0	N/A	N/A	68	44	0	36	8	3	5	20	33.3333333	2.2	N/A	N/A	N/A
Val_73	tumor and benign	low	6	3	3	2b	0	x	0	N/A	N/A	65	5.5	24	60	4	2	2	37	6.6666667	0.14864865	N/A	N/A	N/A
Val_74	tumor and benign	low	7	3	4	2b	0	x	1	N/A	N/A	61	4.7	9	30	7	3	4	26	40	0.18076923	N/A	N/A	N/A
Val_75	tumor and benign	low	7	3	4	2c	x	x	0	N/A	N/A	55	3.15	12		6	3	3	35	20	0.09	N/A	N/A	N/A
Val_76	tumor and benign	low	6	3	3	3a	0	x	0	N/A	N/A	69	6.8	16		7	3	4	26	20	0.26153846	N/A	N/A	N/A
Val_77	tumor and benign	high	8	4	4	2b	0	x	0	N/A	N/A	55	7.4	8		7	3	4	25	36.3636364	0.296	N/A	N/A	N/A
Val_78	tumor and benign	high	9	5	4	2c	x	x	0	N/A	N/A	51	6.9	5	37	7	3	4	25	13.3333333	0.276	N/A	N/A	N/A
Val_79	tumor and benign	high	8	3	5	2c	x	x	0	N/A	N/A	59	4.4	10	50	6	3	3	50	10	0.088	N/A	N/A	N/A

Supplementary Table 2: Detailed information for gene specific Taqman and siRNA assays.

Gene Symbol	Taqman Assay ID	RefSeq	Exon Boundary	Median Cp Tissues Tumor / Benign	Median Cp Cell line PC3 /LNCaP	Dharmacon ON- TARGETplus SMARTpool
MELK	Hs00207681_m1	NM_014791	5-6	38.12 / 40.00	30.70 / 30.54	L-004029-00
UBE2C	Hs00964100_g1	NM_181799.1	5-6	34.01 / 34.58	24.70 / 26.36	L-004693-00
CDC20	Hs00415851_g1	NM_001255.2	1-2	36.26 / 36.71	29.75 / 29.75	N/A
CDCA8	Hs00983655_m1	NM_018101.2	3-4	35.05 / 35.65	28.99 / 28.71	N/A
PRC1	Hs00187740_m1	NM_199413.1	1-2	34.81 / 35.12	N/A	N/A
CCNB2	Hs00270424_m1	NM_004701.2	1-2	32.93 / 33.14	N/A	N/A
POLR2A	Hs00172187_m1	NM_000937.4	1-2	34.68 / 34.60	N/A	N/A
FOXM1	Hs01073586_m1	NM_001243088.1	4-5	N/A	27.51 / 28.01	N/A
B2M	Hs99999907_m1	NM_004048.2	2-3	23.74 / 23.84	23.04 / 24.89	N/A

331	ILMN_1708414	9.2504	0.04676	0.7309	-4.6555	0.4581	GNL3L	54552	331	ILMN_1691143	7.2665	0.04569	1.3376	4.6135	0.4699	RAB18	22931
332	ILMN_3289262	9.2384	0.04676	1.3295	4.6581	0.4628	LOC100131261	100131261	332	ILMN_3289730	9.0697	0.04569	1.4096	4.6106	0.4646	LOC100132199	100132199
333	ILMN_1702447	11.0031	0.04692	0.7329	-4.6520	0.4519	IGF2BP2	10644	333	ILMN_1660858	8.4163	0.04592	0.7460	-4.6057	0.4555	RIN1	9610
334	ILMN_2075927	8.9237	0.04751	0.7719	-4.6441	0.4375	STK40	83931	334	ILMN_1726516	10.3963	0.04592	0.7486	-4.6051	0.4543	SCRIB	23513
335	ILMN_2157277	12.1528	0.04768	0.7698	-4.6397	0.4297	AKR1D1	6718	335	ILMN_1859946	11.8941	0.04611	1.3573	4.6015	0.4478	NA	NA
336	ILMN_1699503	9.2364	0.04768	1.2967	4.6392	0.4287	UBE2D2	7322	336	ILMN_1671911	10.2370	0.04617	0.7040	-4.5994	0.4438	MTA1	9112
337	ILMN_3279414	9.4371	0.04791	1.4390	4.6352	0.4215	LOC440595	440595	337	ILMN_1857017	9.0698	0.04637	0.6896	-4.5955	0.4366	NA	NA
338	ILMN_1669268	8.3261	0.04802	0.7260	-4.6298	0.4118	MEK3D	399664	338	ILMN_3238137	8.0105	0.04637	1.3578	4.5944	0.4345	LOC100133008	100133008
339	ILMN_2044832	10.0503	0.04802	0.7632	-4.6322	0.4162	NOP56	10528	339	ILMN_2371169	11.5712	0.04660	0.6754	-4.5745	0.3979	ZYX	7791
340	ILMN_1732688	9.9642	0.04802	1.3122	4.6295	0.4113	DUT	1854	340	ILMN_1698419	11.0778	0.04660	0.6766	-4.5811	0.4100	NCOR2	9612
341	ILMN_1694100	7.3032	0.04843	1.3229	4.6228	0.3990	PRIM2	5558	341	ILMN_1810055	9.2484	0.04660	0.6787	-4.5787	0.4056	ITFG3	83986
342	ILMN_2151441	8.7502	0.04843	1.3309	4.6222	0.3980	FAM103A1	83640	342	ILMN_1718610	9.1804	0.04660	0.7316	-4.5824	0.4125	ARHGAP17	55114
343	ILMN_2100458	8.3742	0.04847	1.2987	4.6203	0.3945	RFESD	317671	343	ILMN_1663090	7.9010	0.04660	0.7478	-4.5827	0.4131	SON	6651
344	ILMN_2363658	10.0153	0.04851	0.6576	-4.6168	0.3883	PXDN	7837	344	ILMN_1786396	7.7886	0.04660	0.7480	-4.5777	0.4037	ZZEF1	23140
345	ILMN_1764158	7.8799	0.04851	0.7603	-4.6174	0.3893	NLGN2	57555	345	ILMN_1672504	10.6517	0.04660	0.7527	-4.5896	0.4257	PDXK	8566
346	ILMN_3245869	10.9726	0.04851	1.2970	4.6154	0.3857	LOC440957	440957	346	ILMN_1766657	8.1794	0.04660	1.3302	4.5826	0.4128	STOM	2040
347	ILMN_1770892	11.3095	0.04852	0.6998	-4.6120	0.3796	YY1	7528	347	ILMN_3227604	7.6462	0.04660	1.3348	4.5751	0.3990	LOC729342	729342
348	ILMN_1764861	10.1593	0.04852	1.3348	4.6112	0.3781	ISOC1	51015	348	ILMN_1790973	9.1750	0.04660	1.3358	4.5830	0.4135	CDS2	8760
349	ILMN_2246083	7.6871	0.04852	1.4306	4.6108	0.3774	C7orf28B	221960	349	ILMN_3280496	8.1770	0.04660	1.3507	4.5857	0.4186	LOC100131526	100131526
350	ILMN_2049021	12.0396	0.04886	1.3028	4.6058	0.3683	PTTG3P	26255	350	ILMN_3245517	11.0125	0.04660	1.4015	4.5759	0.4004	LOC100134273	100134273
351	ILMN_2169839	10.9808	0.04922	1.3018	4.6006	0.3589	CNPB	7555	351	ILMN_1706521	11.1663	0.04689	0.7406	-4.5700	0.3895	CSNK1G2	1455
352	ILMN_1745034	8.9963	0.04925	1.2765	4.5988	0.3556	SLC11A2	4891	352	ILMN_1667893	10.9722	0.04756	0.7495	-4.5615	0.3738	TNS3	64759
353	ILMN_1806486	8.9830	0.04927	1.2998	4.5972	0.3527	LOC389137	389137	353	ILMN_1743275	8.2612	0.04759	0.7089	-4.5528	0.3577	SH3RF3	344558
354	ILMN_2329735	7.4059	0.04953	0.7689	-4.5930	0.3451	ECM1	1893	354	ILMN_1771987	10.4964	0.04759	0.7417	-4.5557	0.3630	SLC44A2	57153
355	ILMN_1668052	12.3307	0.04969	0.7764	-4.5900	0.3397	FOXA2	3170	355	ILMN_1732151	11.4275	0.04759	0.7528	-4.5545	0.3609	COL6A1	1291
356	ILMN_1652215	9.3913	0.04995	1.4202	4.5858	0.3321	LOC644310	644310	356	ILMN_2252136	10.4406	0.04759	1.3415	4.5533	0.3566	YWHAE	7531
									357	ILMN_1751760	7.4410	0.04759	1.3751	4.5555	0.3627	BCAS2	10286
									358	ILMN_3239574	8.4694	0.04759	1.4702	4.5585	0.3682	SNORD3A	780851
									359	ILMN_2128293	9.1586	0.04790	1.4150	4.5482	0.3491	APIP	51074
									360	ILMN_1706583	9.6275	0.04810	1.3655	4.5439	0.3411	DLAT	1737
									361	ILMN_1701854	9.8411	0.04810	1.3681	4.5434	0.3402	NGF5	2787
									362	ILMN_1766981	10.4598	0.04812	1.3221	4.5419	0.3374	UNC50	25972
									363	ILMN_1810289	11.3773	0.04875	0.7366	-4.5329	0.3208	FER1L3	26509
									364	ILMN_3284177	8.4639	0.04875	1.4501	4.5327	0.3203	LOC100132425	100132425
									365	ILMN_1719985	10.5579	0.04880	0.7294	-4.5307	0.3167	FEM1A	55527
									366	ILMN_1683313	11.1708	0.04904	0.7015	-4.5243	0.3048	ST3GAL1	6482
									367	ILMN_2412380	8.4736	0.04904	1.3691	4.5254	0.3069	TSC22D1	8848
									368	ILMN_3188076	11.6037	0.04904	1.6150	4.5243	0.3047	LOC100128060	100128060
									369	ILMN_1667034	10.2074	0.04923	0.7412	-4.5210	0.2987	PDPR	55066
									370	ILMN_1738093	9.2028	0.04929	0.7566	-4.5191	0.2952	TMEM118	84900
									371	ILMN_1703246	8.6245	0.04947	0.7162	-4.5120	0.2819	SBF1	6305
									372	ILMN_1698225	7.8815	0.04947	0.7350	-4.5141	0.2858	MYO5A	4644
									373	ILMN_2349393	11.5172	0.04947	0.7455	-4.5121	0.2822	MDK	4192
									374	ILMN_2347044	7.9814	0.04947	1.3192	4.5133	0.2844	SLC25A14	9016
									375	ILMN_2410924	8.5868	0.04953	1.4511	4.5100	0.2783	PLOD2	5352
									376	ILMN_3216794	7.5503	0.04955	1.3478	4.5080	0.2745	LOC644860	644860
									377	ILMN_2149400	7.7792	0.04955	1.3741	4.5072	0.2731	SPC25	57405
									378	ILMN_1660793	9.1601	0.04975	0.7434	-4.5026	0.2645	PAQR4	124222
									379	ILMN_1692790	8.6737	0.04975	1.3378	4.5027	0.2648	ITGB3BP	23421

Supplementary Table 5: Downstream processes assessed to the downregulated genes upon MELK and UBE2C RNAi-based knockdown in PC3 or overlapping genes between both experiments.

MELK knockdown deregulated genes

Gene regulation	GO ID	GO Name	Genes	Group count	Total count	p-value
Down	GO:0016568	chromatin modification	carm1; crebbp; setd2; trrap; myst4; arid1a; chd8; smarcc2; smarca4	9	219	2.10E-05
Down	GO:0009790	embryonic development	lama5; slit2; foxc1; fbn2; vegfc; lrp5; lama3; chd8; smarca4	9	235	3.42E-05
Down	GO:0016573	histone acetylation;	myst4; trrap; crebbp	3	13	0.000454
Down	GO:0016477	cell migration	lama5; lrp5; vegfc; lama3; nr2f1; slit2; foxc1	7	233	0.00104

UBE2C knockdown deregulated genes

Gene regulation	GO ID	GO Name	Genes	Group count	Total count	p-value
Down	GO:0016568	chromatin modification	carm1; trrap; ep400; smarcc2; fbxl11; smarca4; setd2; setd1a	8	219	0.000969
Down	GO:0030029	actin filament-based process	abl1; myo5a; c14orf173; cdc42bbp; actn4; myo9b; cdc42bpa; myh9; sptan1	9	260	0.000583
Down	GO:0007067	mitosis	aspm; clip1; shc1; cdc2l1; cenpe; numa1; ube2c; zzef1	8	239	0.00149
Down	GO:0007155	cell adhesion	clstn1; itga3; lama5; abl1; col6a2; inpp1; zyx; itgb4; ptpm; sirpa; lpp; ptpfr; col6a1; cd99	14	960	0.00317
Down	GO:0022403	cell cycle phase	aspm; abl1; clip1; shc1; cdc2l1; cenpe; numa1; ube2c; zzef1	9	369	0.00317

Overlapping genes upon MELK or UBE2C knockdown

Gene regulation	GO ID	GO Name	Genes	Group count	Total count	p-value
Down	GO:0016568	chromatin modification	carm1; trrap; smarcc2; setd2; smarca4	5	219	0.00788
Down	GO:0009790	embryonic development	lrp5; lama5; slit2; smarca4; fbn2	5	235	0.00837

ILMN_1698404	8.8581	7.80E-008	2.3274	10.0461	12.3801	ERN1	2081
ILMN_1679041	7.9585	8.97E-008	2.4060	9.9601	12.2196	SLC3A2	6520
ILMN_1813100	9.6560	1.24E-007	2.0469	9.7525	11.8285	KIAA1244	57221
ILMN_3274596	10.1242	1.47E-007	2.4753	9.6482	11.6299	LOC286512	286512
ILMN_1753342	12.1394	1.64E-007	2.1750	9.5790	11.4973	11/01/11	6303
ILMN_1796069	9.0496	1.72E-007	0.4092	-9.5510	11.4434	CBLN2	147381
ILMN_1673769	8.2937	2.20E-007	2.0272	9.3975	11.1468	KCNG1	3755
ILMN_2052208	8.8649	2.43E-007	2.0672	9.3349	11.0250	GADD45A	1647
ILMN_1696407	11.6204	4.46E-007	0.4506	-8.9675	10.2991	SFRS2	6427
ILMN_1740426	8.3846	4.49E-006	2.3401	7.6320	7.5094	RASD1	51655
ILMN_1881909	12.5125	1.52E-005	0.3338	-6.9841	6.0703		NA
ILMN_1791123	11.3612	0.000137722	2.1707	5.8616	3.4558	TMPRSS2	7113
ILMN_2412336	7.6927	0.000141601	2.0704	5.8475	3.4223	AKR1C2	1646

Supplementary Table 7: Downstream processes assessed to the up- and downregulated genes upon siomycin A treatment.

Siomycin A deregulated genes in LNCaP

Gene regulation	GO ID	GO Name	Genes	Group count	Total count	p-value
Down	GO:0007049	cell cycle	ccnf; mcm6; prc1 ; kpna2; tpx2; kif11; aurka ; c11orf82; h2afx; chek1; kifc1; e2f2 ; mcm3; cdkn3; cdca3; fanci; suv39h1; cdc20 ; ndc80; cdca2; racgap1; cdc25a; cks2; cdc45l ; cdk2; anln; birc5; cdca5; gmnn; spc24; zwint; cdc2; nusap1 ; uhrf1 ; cep55; kntc1 ; exo1; cdc7; bub1; smc4; stmn1 ; ccna2; mns1; cdca8 ; mcm2; pbk; ttk; tubg1; ccnb2 ; cdt1 ; aurkb ; ube2c ; fbxo5; mad2l1; cks1b; ncapg; ccne2; mcm7	58	839	1.45E-57
Down	GO:0006260	DNA replication	orc6l; mcm6; pole3; rfc4; tk1 ; tyms; exo1; mcm3; gins2; mcm4 ; pole2; rfc5; pcna; mcm2; mcm5; maseh2a; cdc45l; cdt1; cdk2; rrm2; top2a ; mcm10; fen1; gmnn; pola2; polq; mcm7; ccne2	28	355	5.30E-28
Down	GO:0007051	spindle organization and biogenesis	prc1; kif11; ndc80; aurka; ttk; tubg1; cks2; stmn1; ube2c; zwint	10	19	6.63E-19
Down	GO:0006281	DNA repair	pcna; fanci; rfc4; tyms; h2afx; chek1; exo1; nudt1; top2a; fen1; polq; pole2; rad51ap1; rfc5; uhrf1	15	392	1.85E-10
Down	GO:0000079	regulation of cyclin-dependent protein kinase activity;	cdkn3; ccna2; cks1b; cdc25a; chek1; ccne2; cks2	7	49	6.18E-09
Down	GO:0008283	cell proliferation	pcna; tpx2; racgap1; ttk; cdc25a; chek1; bub1; cdc7; cks2; cdk2; cdkn3; cks1b; cdca7; uhrf1	14	745	3.77E-06
Up	GO:0006915	apoptosis	trib3; nupr1; cbx4; ern1; ddit3; dnase2; gadd45a; ddit4; cebpg; cebpb	10	855	0.000327

Siomycin A deregulated genes in PC3

Gene regulation	GO ID	GO Name	Genes	Group count	Total count	p-value
Down	GO:0045859	regulation of protein kinase activity	spred1; c5; fabp4; spry1; cdkn2b	5	237	0.00323
Down	GO:0009611	response to wounding	il1b; c5; fabp4; hoxb13; f2rl1; thbs1	6	423	0.00323
Up	GO:0006915	apoptosis	il12a; angptl4; rhob; ddit3; ern1; sqstm1; tgm2; gadd45a; ddit4; blid; pim1; hmox1	12	855	3.18E-05
Up	GO:0008283	cell proliferation	il12a; myc; insig1; adm; fth1; adamts1; tgm2; pim1; cyr61; hmox1	10	745	0.000217
Up	GO:0007050	cell cycle arrest	ddit3; ern1; myc; gadd45a	4	101	0.00202
Up	GO:0008202	steroid metabolic process	akr1c4; hmgs1; insig1; npc1; adm	5	223	0.00289
Up	GO:0048514	blood vessel morphogenesis	angptl4; tgm2; rhob; hmox1	4	147	0.00492