Table S1. Patient characteristics of the 26 AML cases over-expressing *EVI1*

Abbreviations; FAB; French-American British classification, WBC; white blood cell count in 10^9/L.

Patient	Age	Gender	FAB	WBC	HOVON	Karyotype
3102	NA	M	M2	15	NA	45,XY,-7[33]/46,XY[8]
2186	40	M	M5	16.1	HO04	45,XY,-7,t(7;8)(q22;p11)[21]
2250	19	M	M1	9.8	HO29	46,XY,-7,ish add(12)(p13),(wcp12-),+r[13]/46,XY[15]
2276	57	F	M0	35.3	HO29	46,XX,t(1;6)(p32;q24or25),del(2)(q34)[33]46,XX[1], inv(3)(q21q26)*
2327	54	F	M2	29.8	HO04	46,XX,del(7)(q22)[41],46,XX[1]
2207	31	M	M1	96	HO04	46,XY,t(6;11)(q25;q23)
2288	31	M	M4	17.4	HO29	45,XY,-7,t(9;11)(p21;q23)[33]
2664	34	M	M0	173	HO29	45,XY,inv(3)(q12q26.2),-7
2666	36	M	M5	23	HO29	NA
6238	30	F	M4	4.5	HO42	46,XX,t(6;11)(q27;q23)[28]
5288	65	M	M2	25	HO43	45,XY,inv(3)(q21q26)*,-7[16]/46,XY[9]
5291	67	M	Mx	25	HO43	53,XY,+6,+8,+9,t(11;16)(q23;p13),+13,+14,+19,+21
5351	67	F	M4	122	HO43	46,XX,t(6;11)(q27;q23)[36]
5354	72	M	M9	54	HO43	46,XY,t(3;21)(q26;q22),del(12)(p12p13)[20]
6357	61	M	M1	19.7	HO43	45,XY,inv(3)(q21q26)*,-7[16]/46,XY[8]
7072	61	M	M4	23.5	HO42	46,XY,?11(q2?)[3]/46,XY[18] ish t(9;11)(p22;q23)
7084	65	F	M4	24.9	HO43	NA
7306	51	M	M5	33	HO42	46,XY,t(11;19)(q23;p13.1)[15]/46,XY[5]
7416	56	M	M4	20.7	HO42	48,XY,+9,+21[9]/49,XY,+9,+21,+21[14]
6945	33	M	M4	90	HO04	46,XY/47,XY,+?8[2]/48,XY,+mar1,+mar2[28]
3328	41	F	M5	25	HO42	46,XX,t(11;19)(q23;p13)[21]
7136	69	M	M6	11	HO43	45,XY,t(3;3)(q21;q26),-7
2228	41	F	M4	38	HO29	46,XX[66],inv(3)(q21q26)*
2326	31	M	M2	94	HO29	46,XY,t(9;22)(q34;q11)[22]
2190	16	M	M5	54.7	HO04	45,XY,inv(3)(q22q26),-7[25]
2747	23	F	M5	44.8	HO29	45,XX,inv(3)(q2?1q26),-7[31]/46,XX[1]

Table S2. The 294 differentially methylated genes in *EVI1* AML compared to CD34+ normal bone marrow (NBM) (303 probe sets)

The genes are ranked on chromosome location. Methylation difference (mean^{EVI1} log (HpaII/MspI) – mean^{NBM} log (HpaII/MspI) ratio), moderated T-test P-value comparing both groups (BH correct P<0.05). The previously described tumor suppressor genes are colored in red.

HELP probe sets	Chr	RefSeq ID	Gene Symbol	P-value	Meth Diff	Mean EVI1	Mean NBM
MSPI0406S00011571	chr1	NM_138346	KIAA2013	0.0000	2.13	2.04	-0.08
MSPI0406S00020478	chr1	NM_001102398, NM_005826	HNRPR, HNRNPR	0.0000	-1.76	-1.26	0.50
MSPI0406S00028118	chr1			0.0000	-2.12	-0.11	2.01
MSPI0406S00032481	chr1	NM_001905	CTPS	0.0000	-1.58	0.44	2.01
MSPI0406S00033467	chr1	NM_006824, NM_152498	EBNA1BP2, WDR65	0.0001	-1.85	0.90	2.75
MSPI0406S00033551	chr1	NM_005424	TIE1	0.0000	-1.79	0.64	2.42
MSPI0406S00035767	chr1	NM_178033	CYP4X1	0.0001	-1.83	-0.43	1.40
MSPI0406S00035977	chr1	NM_004474	FOXD2	0.0006	-1.52	0.41	1.94
MSPI0406S00047441	chr1	NM_014839	LPPR4	0.0000	-1.88	-1.32	0.56
MSPI0406S00054201	chr1	NM_181873	MTMR11	0.0002	1.63	2.23	0.60
MSPI0406S00054412	chr1	NM_144697	C1orf51	0.0008	-1.76	1.22	2.98
MSPI0406S00054495	chr1	NM_015203	KIAA0460	0.0002	-2.25	-0.23	2.01
MSPI0406S00055568	chr1	NM_016178, NM_031420	OAZ3, MRPL9	0.0000	-2.11	-0.51	1.59
MSPI0406S00055659	chr1	NM_053055	THEM4	0.0000	-2.96	-0.32	2.63
MSPI0406S00055835	chr1	NM_178435	LCE3E	0.0002	2.12	0.87	-1.24
MSPI0406S00055975	chr1	NM_002965	S100A9	0.0006	1.59	0.90	-0.69
MSPI0406S00059603	chr1	NM_004833	AIM2	0.0009	2.27	0.44	-1.83
MSPI0406S00059781	chr1	NM_012337	CCDC19	0.0000	-2.12	0.15	2.28
MSPI0406S00059782	chr1	NM_012337	CCDC19	0.0001	-2.06	0.52	2.58
MSPI0406S00060117	chr1	NM_001778	CD48	0.0009	1.65	0.77	-0.88
MSPI0406S00062872	chr1	NM_015569	DNM3	0.0000	-2.49	0.70	3.18
MSPI0406S00068091	chr1	NM_012482	ZNF281	0.0000	-2.86	-0.61	2.25
MSPI0406S00069595	chr1	NM_006618	JARID1B	0.0000	-2.02	-0.17	1.85
MSPI0406S00074411	chr1	NM_005401	PTPN14	0.0000	-1.91	0.33	2.24
MSPI0406S00099987	chr2	NM_173853	KRTCAP3	0.0000	-1.60	-0.31	1.29
MSPI0406S00103855	chr2	BC030087	LOC375196	0.0000	-1.70	0.79	2.49
MSPI0406S00103856	chr2	BC030087	LOC375196	0.0000	-1.66	-0.94	0.73
MSPI0406S00105608	chr2	NM_024766, NM_001042386	C2orf34, PREPL	0.0000	-2.14	-1.80	0.35
MSPI0406S00107352	chr2	NM_000233	LHCGR	0.0009	-1.74	-0.81	0.93
MSPI0406S00108560	chr2	NM_033109	PNPT1	0.0007	-1.61	-0.45	1.16
MSPI0406S00114344	chr2	NM_001009812	LBX2	0.0000	-1.84	0.68	2.52
MSPI0406S00116274	chr2	NM_006634	VAMP5	0.0000	-1.55	0.53	2.09
MSPI0406S00128707	chr2	NM_014553	TFCP2L1	0.0000	1.92	1.54	-0.38
MSPI0406S00130817	chr2	NM_004807	LOC728969	0.0001	-1.94	0.63	2.56
MSPI0406S00133531	chr2	NM_012233	RAB3GAP1	0.0004	1.52	-0.38	-1.90
MSPI0406S00140656	chr2	NM_000079	CHRNA1	0.0008	1.78	-0.57	-2.36
MSPI0406S00145599	chr2	NM_138468	ICA1L	0.0005	-1.72	-0.09	1.63
MSPI0406S00146431	chr2	N25995	NRP2	0.0000	-2.97	-1.67	1.30
MSPI0406S00156924	chr2	NM_001485	GBX2	0.0000	1.85	3.31	1.46
MSPI0406S00161233	chr2	NM_014808	FARP2	0.0000	-1.94	0.50	2.44
MSPI0406S00166143	chr3	NM_025265	TSEN2	0.0001	1.54	1.53	-0.01

HELP probe sets	Chr	RefSeq ID	Gene Symbol	P-value	Meth Diff	Mean EVI1	Mean NBM
MSPI0406S00174112	chr3	NM_207404	ZNF662	0.0000	-1.87	-1.43	0.43
MSPI0406S00178801	chr3	NM_145071	CISH	0.0001	-2.26	0.86	3.11
MSPI0406S00184632	chr3	NM_001005527	FAM19A4	0.0001	-1.74	0.01	1.75
MSPI0406S00191987	chr3	NM_033364	C3orf15	0.0005	-1.53	1.12	2.65
MSPI0406S00192187	chr3	NM_007085	FSTL1	0.0001	-1.85	0.09	1.94
MSPI0406S00196108	chr3	NM_020741	KIAA1257	0.0002	-1.66	0.58	2.24
MSPI0406S00196163	chr3	NM_000174	GP9	0.0000	-1.73	-1.14	0.58
MSPI0406S00196352	chr3	NM_020187	C3orf37	0.0000	-1.78	0.34	2.13
MSPI0406S00197620	chr3	NM_021203	SRPRB	0.0006	-1.51	-0.80	0.71
MSPI0406S00197824	chr3	NM_001005861	RYK	0.0000	1.68	1.19	-0.49
MSPI0406S00201342	chr3	NM_002628	PFN2	0.0000	-2.40	-0.85	1.55
MSPI0406S00202811	chr3	NM_001099777	C3orf55	0.0001	-1.69	0.69	2.38
MSPI0406S00208333	chr3	NM_144635	FAM131A	0.0000	-1.68	-1.19	0.48
MSPI0406S00213790	chr3	NM_025163	PIGZ	0.0000	-2.37	-1.74	0.63
MSPI0406S00213791	chr3	NM_025163	PIGZ	0.0000	-3.20	-1.65	1.55
MSPI0406S00218942	chr4	NM_016930	STX18	0.0001	-1.77	1.58	3.35
MSPI0406S00220627	chr4	NM_153376, NM_152293	CCDC96, MGC21874	0.0000	-2.04	-1.15	0.89
MSPI0406S00224622	chr4	AB033102	KIAA1276	0.0003	3.07	0.76	-2.31
MSPI0406S00239258	chr4	NM_005908	MANBA	0.0000	-1.70	-0.70	1.00
MSPI0406S00239368	chr4	NM_001059	TACR3	0.0001	1.85	0.50	-1.35
MSPI0406S00240343	chr4	NM_024090	ELOVL6	0.0008	1.76	2.10	0.33
MSPI0406S00245845	chr4	NM_004575	POU4F2	0.0000	1.76	2.46	0.71
MSPI0406S00247238	chr4	NM_015271	TRIM2	0.0003	-1.62	-1.56	0.06
MSPI0406S00247456	chr4	NM_003264	TLR2	0.0002	-2.13	0.03	2.16
MSPI0406S00247772	chr4	NM_000857	GUCY1B3	0.0000	-2.06	-0.54	1.52
MSPI0406S00265126	chr5			0.0000	-2.58	-1.07	1.52
MSPI0406S00265127	chr5			0.0000	-1.54	1.35	2.90
MSPI0406S00265663	chr5	NM_016107	ZFR	0.0003	-2.10	0.01	2.11
MSPI0406S00268436	chr5	NM_021072	HCN1	0.0008	-1.65	-1.53	0.12
MSPI0406S00269033	chr5	NM_019087	ARL15	0.0005	-1.62	0.45	2.07
MSPI0406S00270454	chr5	NM_001048249	DKFZP686E2158	0.0000	-1.82	-0.33	1.49
MSPI0406S00270455	chr5	NM_001048249	DKFZP686E2158	0.0000	-2.26	-0.42	1.84
MSPI0406S00274414	chr5	NM_001882	CRHBP	0.0000	-3.29	-0.71	2.57
MSPI0406S00275324	chr5	NM_003248	THBS4	0.0003	-2.11	-0.12	2.00
MSPI0406S00276902	chr5	NM_004365	CETN3	0.0000	-2.01	0.04	2.04
MSPI0406S00284948	chr5	NM_006161	NEUROG1	0.0000	-2.29	-1.43	0.86
MSPI0406S00286279	chr5	NM_018834	MATR3	0.0001	-2.38	-1.43	0.96
MSPI0406S00287342	chr5	NM_020957, NM_019119	PCDHB16, PCDHB9	0.0003	-1.66	1.10	2.76
MSPI0406S00288076	chr5	AK024556		0.0005	1.82	1.23	-0.59
MSPI0406S00288561	chr5	NM_021182	HMHB1	0.0000	-1.53	-0.52	1.01
MSPI0406S00304193	chr6			0.0000	-1.64	0.54	2.18
MSPI0406S00305367	chr6	NM_012135	FAM50B	0.0000	-1.64	-0.08	1.55
MSPI0406S00310640	chr6	NM_153020	RBM24	0.0001	-2.13	0.40	2.53
MSPI0406S00312897	chr6	AB002384	C6orf32	0.0001	-1.61	0.64	2.25
MSPI0406S00314948	chr6	M80469, AI933930	LOC731974, C6orf12	0.0000	-1.53	1.00	2.53
MSPI0406S00315716	chr6	NM_002701	POU5F1P4	0.0000	-2.28	-1.70	0.58
MSPI0406S00315919	chr6	NM_130463, NM_005007	ATP6V1G2, NFKBIL1	0.0000	-1.63	-0.16	1.46
MSPI0406S00315947	chr6	NM_000594	TNF	0.0008	-1.58	0.25	1.82
MSPI0406S00316051	chr6	NM_021184	C6orf47	0.0000	1.50	2.75	1.25

HELP probe sets	Chr	RefSeq ID	Gene Symbol	P-value	Meth Diff	Mean EVI1	Mean NBM
MSPI0406S00317719	chr6	NM_181336	LEMD2	0.0000	-2.05	-0.39	1.66
MSPI0406S00318806	chr6	NM_001093728	TCP11	0.0004	1.51	1.14	-0.38
MSPI0406S00321358	chr6	NM_015345	DAAM2	0.0002	-1.87	-1.15	0.72
MSPI0406S00323258	chr6	NM_015089	PARC	0.0000	-2.15	-1.33	0.83
MSPI0406S00325106	chr6	NM_153344	C6orf141	0.0007	-1.85	0.56	2.40
MSPI0406S00326033	chr6	NM_001010872	FAM83B	0.0000	-1.93	0.78	2.71
MSPI0406S00326173	chr6	NM_030820	COL21A1	0.0006	-2.23	0.74	2.97
MSPI0406S00334234	chr6	NM_173672, NM_003080	PPIL6, SMPD2	0.0001	-1.69	-1.39	0.29
MSPI0406S00334364	chr6	NM_005284	GPR6	0.0001	-1.65	1.24	2.89
MSPI0406S00337228	chr6	NM_001105545	ECHDC1	0.0002	1.51	3.81	2.31
MSPI0406S00348529	chr6	NM_175922	PRR18	0.0003	-2.00	-0.31	1.70
MSPI0406S00354656	chr7	NM_001013836	MAD1L1	0.0004	-1.63	0.44	2.07
MSPI0406S00363079	chr7	NM_005522	HOXA1	0.0001	-2.05	0.93	2.98
MSPI0406S00363085	chr7	NM_006735	HOXA2	0.0003	-1.81	1.04	2.84
MSPI0406S00363385	chr7	NM_152740	HIBADH	0.0001	-1.55	0.24	1.79
MSPI0406S00368119	chr7	NM_001129	AEBP1	0.0000	-1.83	-0.17	1.66
MSPI0406S00372326	chr7	NM_016139	CHCHD2	0.0000	-1.77	-0.11	1.66
MSPI0406S00394211	chr7	NM_013332	HIG2	0.0000	-1.94	0.52	2.46
MSPI0406S00394642	chr7	NM_012470	TNPO3	0.0000	-2.03	-1.18	0.85
MSPI0406S00400025	chr7	NM_032982	CASP2	0.0000	-1.83	-0.40	1.42
MSPI0406S00413619	chr8	NM_024607	PPP1R3B	0.0000	3.52	2.18	-1.35
MSPI0406S00416381	chr8	NM_019851	FGF20	0.0002	-1.93	1.32	3.24
MSPI0406S00417217	chr8	NM_018371	ChGn	0.0008	1.54	0.80	-0.74
MSPI0406S00418096	chr8	NM_182795	NPM2	0.0001	-2.07	0.26	2.32
MSPI0406S00418452	chr8	NM_015359	SLC39A14	0.0000	-2.90	-0.19	2.72
MSPI0406S00419336	chr8	NM_002318	LOXL2	0.0005	-1.57	1.19	2.76
MSPI0406S00419479	chr8	NM_006167	NKX3-1	0.0001	-2.26	-0.10	2.16
MSPI0406S00423672	chr8	NM_007198	PROSC	0.0000	-2.13	-1.24	0.89
MSPI0406S00423826	chr8	NM_001002814	RAB11FIP1	0.0007	1.56	0.05	-1.51
MSPI0406S00425232	chr8	NM_178819	AGPAT6	0.0000	-2.08	-0.78	1.31
MSPI0406S00429579	chr8	NM_138969	RDHE2	0.0002	-1.51	-2.60	-1.08
MSPI0406S00430774	chr8	NM_000370	TTPA	0.0001	-1.51	-1.36	0.15
MSPI0406S00431037	chr8	NM_004820	CYP7B1	0.0000	-1.80	1.09	2.89
MSPI0406S00438678	chr8			0.0001	1.63	1.46	-0.17
MSPI0406S00456924	chr9	NM_004432	ELAVL2	0.0000	-1.75	1.31	3.05
MSPI0406S00457313	chr9	NM_000459	TEK	0.0000	-2.21	-2.10	0.11
MSPI0406S00457963	chr9	NM_005802	TOPORS	0.0000	-1.71	0.22	1.92
MSPI0406S00459077	chr9	NM_147162	IL11RA	0.0000	-2.97	-0.91	2.06
MSPI0406S00460689	chr9	NM_016042	EXOSC3	0.0000	-2.24	0.28	2.52
MSPI0406S00460735	chr9	NM_033412	MCART1	0.0001	-2.35	-1.09	1.26
MSPI0406S00466605	chr9	NM_001098802	CEP78	0.0000	-1.82	-1.47	0.35
MSPI0406S00468557	chr9	NM_030940	ISCA1	0.0001	-1.59	0.41	2.01
MSPI0406S00471981	chr9	NM_005392	PHF2	0.0000	-1.56	-0.04	1.52
MSPI0406S00474891	chr9	NM_004612	TGFBR1	0.0000	-1.63	-0.01	1.62
MSPI0406S00476961	chr9	NM_003798	CTNNAL1	0.0000	-1.73	0.06	1.79
MSPI0406S00478537	chr9	NM_001006615	WDR31	0.0002	-1.77	-0.66	1.11
MSPI0406S00488286	chr9	NM_014285	EXOSC2	0.0000	-2.53	0.73	3.26
MSPI0406S00490859	chr9			0.0000	-1.84	-0.02	1.83

HELP probe sets	**Chr**	**RefSeq ID**	**Gene Symbol**	**P-value**	**Meth Diff**	**Mean EVI1**	**Mean NBM**
MSPI0406S00494265	chr9	NM_152833	C9orf69	0.0002	1.91	1.14	-0.77
MSPI0406S00500513	chr10	NM_000417	IL2RA	0.0001	1.75	2.06	0.31
MSPI0406S00501433	chr10	NM_031923	TAF3	0.0000	-2.17	1.58	3.76
MSPI0406S00520388	chr10	NM_018649	H2AFY2	0.0002	-1.70	-0.70	0.99
MSPI0406S00528396	chr10	NM_003972	BTAF1	0.0000	-1.94	-1.11	0.83
MSPI0406S00528891	chr10	NM_183374	CYP26C1	0.0000	-2.64	-0.51	2.13
MSPI0406S00533558	chr10	NM_017787	C10orf26	0.0000	-2.54	-1.62	0.92
MSPI0406S00533880	chr10	NM_032727	INA	0.0005	-1.67	0.21	1.88
MSPI0406S00537146	chr10	NM_001003407	ABLIM1	0.0000	-2.50	-0.04	2.46
MSPI0406S00543249	chr10	NM_207426	FLJ46831	0.0000	-1.52	-0.42	1.10
MSPI0406S00553202	chr11	NM_014469	RBMXL2	0.0000	-1.78	-0.94	0.85
MSPI0406S00557657	chr11	NM_006157	NELL1	0.0003	-2.42	-0.71	1.71
MSPI0406S00558409	chr11	NM_030771	CCDC34	0.0000	-3.04	-2.54	0.50
MSPI0406S00559240	chr11	NM_000378, NM_015855	WT1, WIT1	0.0002	-1.67	2.09	3.76
MSPI0406S00562783	chr11	NM_057174, NM_152312	PEX16, GYLTL1B	0.0000	-1.52	-0.48	1.03
MSPI0406S00564200	chr11	NM_015308	FNBP4	0.0006	1.51	1.58	0.07
MSPI0406S00566280	chr11	NM_004778	GPR44	0.0000	-1.67	-0.01	1.66
MSPI0406S00570572	chr11	NM_145719	TIGD3	0.0000	-3.39	-0.08	3.31
MSPI0406S00576824	chr11	NM_021046	KRTAP5-8	0.0002	2.00	0.21	-1.79
MSPI0406S00577268	chr11	NM_005169	PHOX2A	0.0005	-1.90	0.51	2.41
MSPI0406S00578047	chr11	NM_021200	PLEKHB1	0.0000	-1.51	0.95	2.47
MSPI0406S00579448	chr11	NM_020193	C11orf30	0.0000	1.53	2.48	0.96
MSPI0406S00579815	chr11	NM_000260	MYO7A	0.0001	-1.87	-0.18	1.69
MSPI0406S00582986	chr11	NM_130847	AMOTL1	0.0000	-1.89	0.48	2.37
MSPI0406S00585712	chr11	NM_006235	POU2AF1	0.0001	-2.28	-0.13	2.15
MSPI0406S00586869	chr11	NM_015523	REXO2	0.0001	-1.92	-0.50	1.42
MSPI0406S00588918	chr11	NM_015157	PHLDB1	0.0001	1.90	0.14	-1.76
MSPI0406S00592220	chr11	NM_022062	PKNOX2	0.0009	-1.50	0.48	1.98
MSPI0406S00592482	chr11	NM_005103	FEZ1	0.0001	-1.81	0.88	2.70
MSPI0406S00600078	chr12	NM_000552	VWF	0.0000	-2.03	0.39	2.42
MSPI0406S00600155	chr12	NM_001769	CD9	0.0006	-1.70	1.19	2.90
MSPI0406S00601262	chr12	NM_020634	GDF3	0.0001	-2.15	0.36	2.51
MSPI0406S00602696	chr12	NM_003651	CSDA	0.0000	-2.00	0.23	2.23
MSPI0406S00606787	chr12	NM_006390	IPO8	0.0000	-2.30	-2.06	0.24
MSPI0406S00612781	chr12	NM_000020	ACVRL1	0.0005	-1.52	0.44	1.95
MSPI0406S00613096	chr12	NM_002281	KRT81	0.0000	-2.57	-1.15	1.43
MSPI0406S00614215	chr12	NM_020898	CALCOCO1	0.0000	-1.57	-1.19	0.38
MSPI0406S00614766	chr12	NM_006741	PPP1R1A	0.0000	-1.77	-0.87	0.90
MSPI0406S00616460	chr12	NM_133483, NM_001111270	GEFT	0.0000	1.53	3.87	2.34
MSPI0406S00617895	chr12	NM_001031679	MSRB3	0.0001	-2.26	0.62	2.87
MSPI0406S00619189	chr12	NM_002849	PTPRR	0.0000	-1.80	-1.53	0.28
MSPI0406S00625770	chr12	NM_152261	C12orf23	0.0002	-2.52	-0.38	2.15
MSPI0406S00626032	chr12	NM_012406	PRDM4	0.0000	-1.54	-1.80	-0.26
MSPI0406S00634978	chr12	NM_001024808	BCL7A	0.0007	-1.66	1.02	2.68
MSPI0406S00641583	chr12	NM_003565	ULK1	0.0001	-1.56	-0.34	1.22
MSPI0406S00646456	chr13	NM_001260	CDK8	0.0000	-2.35	-0.75	1.60
MSPI0406S00647008	chr13	NM_002097	GTF3A	0.0010	-2.04	0.51	2.55
MSPI0406S00649799	chr13	NM_203451	LOC400120	0.0000	-1.71	-0.28	1.43

HELP probe sets	Chr	RefSeq ID	Gene Symbol	P-value	Meth Diff	Mean EVI1	Mean NBM
MSPI0406S00652106	chr13	NM_015070	ZC3H13	0.0002	-1.79	0.36	2.15
MSPI0406S00653659	chr13	NM_024705	DHRS12	0.0000	-3.02	-0.75	2.27
MSPI0406S00661059	chr13	NM_033132	ZIC5	0.0004	-1.66	1.91	3.57
MSPI0406S00668987	chr14	NM_004643	PABPN1	0.0000	-2.56	-1.59	0.97
MSPI0406S00669167	chr14	BE673662	ZFHX2	0.0006	-1.86	-0.56	1.30
MSPI0406S00670532	chr14	NM_004086	COCH	0.0000	-1.95	-0.48	1.46
MSPI0406S00676765	chr14	NM_001079520	DACT1	0.0000	-2.07	-0.07	2.00
MSPI0406S00680432	chr14	NM_033262	SLC8A3	0.0001	-1.72	-0.59	1.13
MSPI0406S00685030	chr14	NM_015859	GTF2A1	0.0001	-2.18	0.31	2.49
MSPI0406S00686729	chr14	NM_022054	KCNK13	0.0006	-2.15	-0.35	1.81
MSPI0406S00690443	chr14	NM_018036	ATG2B	0.0000	-1.56	-0.98	0.58
MSPI0406S00696953	chr14	AI935123	AHNAK2	0.0001	1.59	-0.17	-1.76
MSPI0406S00696954	chr14	AI935123	AHNAK2	0.0000	2.01	1.50	-0.50
MSPI0406S00700799	chr15	NM_024490	ATP10A	0.0001	1.99	1.96	-0.03
MSPI0406S00701188	chr15	NM_033223	GABRG3	0.0000	-1.93	-0.10	1.83
MSPI0406S00703260	chr15	NM_017762	MTMR10	0.0000	1.59	1.60	0.01
MSPI0406S00704116	chr15	NM_003020	SCG5	0.0000	-2.78	-1.42	1.36
MSPI0406S00706126	chr15	NM_033286	C15orf23	0.0005	-1.53	1.32	2.85
MSPI0406S00714988	chr15	NM_194272	RBPMS2	0.0000	-1.63	-0.61	1.03
MSPI0406S00717528	chr15	NM_017705	PAQR5	0.0000	-2.33	-0.10	2.22
MSPI0406S00720935	chr15	NM_001897	CSPG4	0.0000	-1.70	0.03	1.74
MSPI0406S00722613	chr15	NM_006791	MORF4L1	0.0010	-1.76	-1.21	0.55
MSPI0406S00723766	chr15	NM_032246	MEX3B	0.0001	-1.82	1.38	3.20
MSPI0406S00726849	chr15			0.0000	-1.79	0.50	2.29
MSPI0406S00735207	chr16	NM_023076	C16orf28	0.0002	-1.76	-0.71	1.05
MSPI0406S00736025	chr16	NM_144603	NOXO1	0.0000	-1.68	0.83	2.51
MSPI0406S00736456	chr16	NM_001374	DNASE1L2	0.0000	-1.56	0.02	1.58
MSPI0406S00745502	chr16	NM_033201	C16orf45	0.0000	-1.85	-0.11	1.74
MSPI0406S00751950	chr16	NM_015202, NM_001520	KIAA0556, GTF3C1	0.0000	-1.96	0.09	2.05
MSPI0406S00754682	chr16	NM_016643	ZNF771	0.0000	-1.62	-1.24	0.39
MSPI0406S00755815	chr16	NM_015927	TGFB1I1	0.0004	-1.64	-0.34	1.30
MSPI0406S00760933	chr16	NM_005953	MT2A	0.0000	-2.03	-1.57	0.46
MSPI0406S00765599	chr16	NM_018667	SMPD3	0.0007	-1.66	0.22	1.88
MSPI0406S00772241	chr16	NM_022041	GAN	0.0002	-1.74	1.24	2.98
MSPI0406S00777297	chr16	NM_015144	ZCCHC14	0.0000	-1.75	-0.65	1.10
MSPI0406S00777570	chr16	AI797462	KLHDC4	0.0005	-1.60	-1.17	0.43
MSPI0406S00778118	chr16			0.0000	-1.79	0.23	2.02
MSPI0406S00785511	chr17			0.0000	3.69	2.16	-1.53
MSPI0406S00785512	chr17			0.0000	2.13	2.33	0.20
MSPI0406S00786244	chr17	NM_153018	ZFP3	0.0000	-1.73	-0.12	1.61
MSPI0406S00787973	chr17	NM_001002914	KCTD11	0.0000	-1.58	-0.14	1.44
MSPI0406S00788425	chr17	NM_001406	EFNB3	0.0000	-1.85	1.09	2.94
MSPI0406S00788661	chr17	NM_004732	KCNAB3	0.0001	-1.64	-1.06	0.58
MSPI0406S00788662	chr17	NM_004732	KCNAB3	0.0000	-1.66	-1.59	0.07
MSPI0406S00788663	chr17	NM_004732, NM_053051, NM_021210	KCNAB3, CNTROB, TRAPPC1	0.0000	-2.97	-1.49	1.48
MSPI0406S00791659	chr17	NM_014859	RICH2	0.0000	-2.83	-1.38	1.45
MSPI0406S00793116	chr17	NM_181716	PRR6	0.0000	-2.32	-1.65	0.67

HELP probe sets	Chr	RefSeq ID	Gene Symbol	P-value	Meth Diff	Mean EVI1	Mean NBM
MSPI0406S00798560	chr17	NM_144610	FLJ25006	0.0000	-2.49	-1.84	0.64
MSPI0406S00801497	chr17	NM_183377	ACCN1	0.0006	-1.53	-1.59	-0.06
MSPI0406S00810245	chr17	NM_174919	LOC201175	0.0005	-2.20	-1.20	0.99
MSPI0406S00811398	chr17	NM_001002841	MYL4	0.0000	-1.87	-1.03	0.84
MSPI0406S00812189	chr17			0.0008	-1.64	0.66	2.31
MSPI0406S00817603	chr17	NM_181707	C17orf64	0.0007	1.82	1.35	-0.47
MSPI0406S00818930	chr17	NM_015623	TANC2	0.0000	-2.11	-0.48	1.63
MSPI0406S00818949	chr17	NM_001017917	CYB561	0.0000	-1.79	0.61	2.40
MSPI0406S00826117	chr17			0.0000	-1.83	-0.53	1.30
MSPI0406S00826166	chr17	NM_001003715, NM_013260	RECQL5, SAP30BP	0.0006	-2.16	-0.04	2.12
MSPI0406S00826327	chr17			0.0000	-2.00	0.39	2.39
MSPI0406S00829958	chr17	NM_138793	CANT1	0.0000	-2.90	-1.30	1.60
MSPI0406S00831072	chr17	BC001525	TBC1D16	0.0002	-2.13	0.45	2.57
MSPI0406S00831073	chr17	BC001525	TBC1D16	0.0008	-1.57	-0.25	1.32
MSPI0406S00838340	chr18	NM_003409	ZFP161	0.0004	-1.57	1.26	2.83
MSPI0406S00841050	chr18	NM_032525	TUBB6	0.0000	-2.29	0.66	2.96
MSPI0406S00841051	chr18	NM_032525	TUBB6	0.0000	-2.21	0.82	3.03
MSPI0406S00844006	chr18	NM_080597	OSBPL1A	0.0006	-1.73	-0.94	0.79
MSPI0406S00847965	chr18	NM_152470	RNF165	0.0000	-1.55	-0.96	0.58
MSPI0406S00848242	chr18	NM_004671	PIAS2	0.0000	-2.87	-0.58	2.29
MSPI0406S00852330	chr18	NM_021127	PMAIP1	0.0000	-3.59	-1.23	2.36
MSPI0406S00858022	chr18	NM_004715	CTDP1	0.0000	1.73	1.37	-0.35
MSPI0406S00861872	chr19	NM_012458	TIMM13	0.0000	-1.66	-0.31	1.35
MSPI0406S00862264	chr19	NM_145173	DIRAS1	0.0004	1.58	1.59	0.01
MSPI0406S00866032	chr19	U40317	PTPRS	0.0000	1.75	2.10	0.35
MSPI0406S00868626	chr19	NM_006702	PNPLA6	0.0001	-1.51	1.41	2.93
MSPI0406S00870600	chr19	NM_152476	ZNF560	0.0003	-2.32	-0.85	1.47
MSPI0406S00874436	chr19	AI937201	NFIX	0.0000	-2.62	1.02	3.64
MSPI0406S00881670	chr19	NM_153221	CILP2	0.0003	-1.73	0.27	2.00
MSPI0406S00890230	chr19	NM_181786	HKR1	0.0008	-1.89	1.21	3.10
MSPI0406S00897488	chr19	NM_030785	RSHL1	0.0004	-1.89	1.51	3.39
MSPI0406S00898094	chr19	NM_020709	KIAA1183	0.0001	-1.50	1.28	2.78
MSPI0406S00900558	chr19	AF245447	SPHK2	0.0000	-1.88	-0.07	1.81
MSPI0406S00900627	chr19	NM_145807	LOC126147	0.0000	-2.13	-1.44	0.69
MSPI0406S00901219	chr19	W67744	SNRP70	0.0003	-1.88	-0.15	1.73
MSPI0406S00902145	chr19	BC024312	MED25	0.0000	1.97	2.90	0.93
MSPI0406S00903958	chr19	AI479923, T81826	LOC339352, FLJ30403	0.0000	-2.60	-0.48	2.13
MSPI0406S00904370	chr19	NM_001010851	ZNF766	0.0000	-1.93	-0.63	1.30
MSPI0406S00908309	chr19	NM_144690	ZNF582	0.0000	-3.31	-3.11	0.19
MSPI0406S00908340	chr19	NM_022103	ZNF667	0.0001	-3.01	-0.45	2.56
MSPI0406S00908349	chr19	NM_020813	ZNF471	0.0007	-1.61	-1.21	0.40
MSPI0406S00908360	chr19	NM_020828	ZFP28	0.0000	-2.59	-0.06	2.53
MSPI0406S00911124	chr20	NM_024325	ZNF343	0.0000	-1.88	0.39	2.27
MSPI0406S00913877	chr20	NM_018848, NM_001009608	MKKS, C20orf94	0.0001	-1.72	0.03	1.75
MSPI0406S00914181	chr20	NM_014962	BTBD3	0.0009	-1.55	1.61	3.16
MSPI0406S00914183	chr20	NM_014962	BTBD3	0.0005	-1.91	1.30	3.21
MSPI0406S00921311	chr20	NM_021202	TP53INP2	0.0000	-1.70	-0.19	1.51
MSPI0406S00923469	chr20	NM_005386	NNAT	0.0004	-1.89	0.15	2.04

HELP probe sets	Chr	RefSeq ID	Gene Symbol	P-value	Meth Diff	Mean EVI1	Mean NBM
MSPI0406S00923470	chr20	NM_005386	NNAT	0.0000	-1.96	-0.13	1.83
MSPI0406S00925914	chr20	NM_001098798	TOX2	0.0001	1.93	0.97	-0.96
MSPI0406S00926213	chr20	NM_178850	HNF4A	0.0000	-2.60	0.69	3.29
MSPI0406S00929615	chr20	NM_000961	PTGIS	0.0009	-1.86	1.00	2.86
MSPI0406S00934116	chr20	NM_080618	CTCFL	0.0010	-1.56	-0.51	1.04
MSPI0406S00934954	chr20	NM_001001433	STX16	0.0000	-1.63	-1.43	0.20
MSPI0406S00946573	chr21	NM_018962	DSCR6	0.0007	-1.95	1.19	3.14
MSPI0406S00951810	chr21	NM_198696	KRTAP10-3	0.0000	-1.81	-1.27	0.54
MSPI0406S00960076	chr22	NM_013378	VPREB3	0.0000	-2.28	-0.66	1.63
MSPI0406S00961868	chr22	NM_021115	SEZ6L	0.0001	1.69	1.34	-0.35
MSPI0406S00970740	chr22	NM_014292	CBX6	0.0007	1.59	0.47	-1.12
MSPI0406S00971904	chr22			0.0002	-1.90	0.93	2.83
MSPI0406S00988001	chrX	NM_015507	EGFL6	0.0007	-1.69	0.30	1.99
MSPI0406S00990104	chrX	NM_152780	MAP7D2	0.0000	-1.77	-0.52	1.25
MSPI0406S00998164	chrX	NM_006150	LMO6	0.0002	-1.52	0.25	1.77
MSPI0406S00998450	chrX	NM_000084	CLCN5	0.0000	-1.70	-0.49	1.21
MSPI0406S00998736	chrX	NM_018094	GSPT2	0.0003	-2.44	-1.65	0.79
MSPI0406S01007247	chrX	NM_080737	SYTL4	0.0000	-1.62	-0.56	1.06
MSPI0406S01007951	chrX	NM_030639	BHLHB9	0.0000	-1.89	-0.49	1.40
MSPI0406S01010836	chrX	NM_173798	ZCCHC12	0.0000	-2.40	-0.31	2.08
MSPI0406S01011804	chrX	NM_014060	MCTS1	0.0004	-2.11	-2.02	0.09
MSPI0406S01020295	chrX	NM_021806	FAM3A	0.0000	-2.49	0.11	2.59

Table S3. Gene Ontology (GO) categories highly represented among the 294 differentially methylated genes

GO Term	# Genes	% Genes	P-Value	Fold Enrichment	FDR (%)
Intracellular membrane-bound organelle	125	43.7%	3.78E-04	1.24	0.57
Regulation of transcription	55	19.2%	1.85E-03	1.47	3.23
Transcription, DNA-dependent	51	17.8%	5.76E-03	1.43	9.73
RNA biosynthetic process	51	17.8%	5.90E-03	1.42	9.95
Cell morphogenesis	16	5.6%	5.38E-03	2.23	9.12
Cellular morphogenesis during differentiation	8	2.8%	7.63E-03	3.52	12.68

FDR; false discovery rate.

Table S4. FIRE analysis on the differentially methylated *EVI1* AML genes and a separate analysis on the hyper- and hypomethylated genes

FIRE analysis differentially methylated genes (n=294)						
Optimized Motif	**Location**	**Robustness**	**Orientation Bias**	**Conservation Index**	**Seed**	**Motif name**
[CGT][AT]CGTA[GT]C[CG]	5'	10/10	←	0.48	ACGTAGC	-
[CGT]CTGACA.[AC]	5'	9/10	←	0.54	CTGACAA	-
[CT]AA[AC]GCC[GT]	5'	8/10	←	0.31	AAAGCCT	-
FIRE analysis *hyper* methylated genes (n=238) and *hypo* methylated genes (n=56)						
Optimized Motif	**Location**	**Robustness**	**Orientation Bias**	**Conservation Index**	**Seed**	**Motif name**
[AG]A[AG]TCATT[GT]	5'	9/10	-	0.79	AATCATT	GCN4
.C[CG]T[GT]ACA[ACT]	5'	9/10	-	0.31	CCTGACA	-
[AC]CCTACC[AC].	5'	6/10	→	0.36	CCTACCC	P
[ACG]AGCGGCG.	5'	7/10	-	0.88	AGCGGCG	-

For each motif, location, i.e. 5′ upstream region, robustness score ranging from 0/10 to 10/10 obtained from ten jack-knife trails of randomly removing one-third of the genes and reassessing the statistical significance, orientation bias indicator, conservation index and seed that gave rise to the motif.

Table S5. The 110 genes differentially methylated between the two *EVI1* subclusters, i.e Cluster A, enriched for *MLL*-rearranged AMLs and Cluster B, enriched for 3q26 aberrations and monosomy 7

HELP probe sets	Chr.	Ref Seq ID	Gene Symbol	P-value	Meth Diff	Mean Cluster A	Mean Cluster B	CpG Cluster	CpG Island
MSPI0406S00014167	chr1	NM_015164	PLEKHM2	0.0000	2.17	2.04	-0.13	-	-
MSPI0406S00014983	chr1	NM_018090, NM_198546	NECAP2, SPATA21	0.0000	1.67	0.67	-1.00	YES	YES
MSPI0406S00019401	chr1	NM_005529	HSPG2	0.0000	1.52	0.97	-0.56	YES	-
MSPI0406S00027314	chr1	NM_175852	TXLNA	0.0009	1.65	1.11	-0.54	YES	-
MSPI0406S00029456	chr1	NM_005202	COL8A2	0.0000	1.51	1.54	0.03	-	-
MSPI0406S00033843	chr1	NM_014663	JMJD2A	0.0001	1.68	0.72	-0.96	YES	-
MSPI0406S00049084	chr1	NM_020703	AMIGO1	0.0000	1.56	1.16	-0.40	YES	YES
MSPI0406S00054187	chr1	NM_014849	SV2A	0.0006	1.72	2.14	0.42	-	-
MSPI0406S00057660	chr1	NM_001105203	RUSC1	0.0000	1.77	2.02	0.26	-	-
MSPI0406S00060281	chr1	NM_001025598	ARHGAP30	0.0000	1.54	0.69	-0.85	-	-
MSPI0406S00062858	chr1	NM_001007239	KIAA0859	0.0001	2.12	0.26	-1.86	YES	YES
MSPI0406S00093425	chr2	NM_002539	ODC1	0.0001	1.71	0.50	-1.21	-	-
MSPI0406S00122762	chr2	NM_001039492	FHL2	0.0001	1.60	1.82	0.21	YES	YES
MSPI0406S00123952	chr2	NM_022336	EDAR	0.0000	1.75	1.04	-0.71	YES	-
MSPI0406S00146170	chr2	NM_152526	PARD3B	0.0009	1.53	2.13	0.61	YES	-
MSPI0406S00161233	chr2	NM_014808	FARP2	0.0000	1.80	1.47	-0.33	YES	-
MSPI0406S00173130	chr3	NM_002295	RPSA	0.0002	1.93	1.79	-0.14	YES	YES
MSPI0406S00180421	chr3	NA		0.0000	1.85	1.08	-0.77	-	-
MSPI0406S00208027	chr3	NM_005688	ABCC5	0.0003	1.60	-0.57	-2.17	YES	-
MSPI0406S00236887	chr4	NM_138980	MAPK10	0.0006	1.59	-0.05	-1.64	-	-
MSPI0406S00238917	chr4	NM_002106	H2AFZ	0.0002	1.55	1.09	-0.46	YES	-
MSPI0406S00245296	chr4	NM_002039	GAB1	0.0002	2.23	1.75	-0.48	YES	YES
MSPI0406S00250036	chr4	NM_016228	AADAT	0.0001	1.68	0.68	-1.00	YES	-
MSPI0406S00260211	chr5	NA		0.0006	2.78	0.39	-2.39	-	-
MSPI0406S00290573	chr5	BE673151	MYOZ3	0.0000	1.59	1.71	0.12	YES	-
MSPI0406S00293417	chr5	NM_022090	LOC63920	0.0001	1.51	-1.14	-2.65	-	-
MSPI0406S00301996	chr5	NM_024978		0.0002	2.44	1.91	-0.52	-	-
MSPI0406S00301997	chr5	NM_024978		0.0005	1.83	1.07	-0.76	-	-
MSPI0406S00302259	chr5	NM_018434	RNF130	0.0001	1.89	2.20	0.31	YES	-
MSPI0406S00316130	chr6	NM_013974	DDAH2	0.0001	1.76	1.99	0.23	YES	YES
MSPI0406S00319826	chr6	NA		0.0006	1.81	1.28	-0.53	-	-
MSPI0406S00346373	chr6	NM_003058	SLC22A2	0.0004	1.58	1.87	0.29	-	-
MSPI0406S00353042	chr7	NM_001505	GPER	0.0000	1.50	0.87	-0.63	YES	YES
MSPI0406S00361726	chr7	NM_000600	IL6	0.0000	1.63	2.55	0.92	-	-
MSPI0406S00363122	chr7	NM_019102	HOXA5	0.0003	2.26	1.39	-0.88	YES	YES
MSPI0406S00368847	chr7	NA		0.0006	1.64	-0.06	-1.70	-	-
MSPI0406S00400041	chr7	NM_000083	CLCN1	0.0000	1.92	0.22	-1.71	-	-
MSPI0406S00400042	chr7	NM_000083	CLCN1	0.0000	2.07	1.28	-0.80	-	-
MSPI0406S00414913	chr8	NM_054029	C8orf14	0.0002	2.07	0.98	-1.09	-	-
MSPI0406S00427034	chr8	NM_001080394	KIAA0146	0.0002	1.78	1.33	-0.46	YES	YES
MSPI0406S00440889	chr8	NM_022783	DEPDC6	0.0000	1.97	2.65	0.68	YES	YES

HELP probe sets	Chr.	Ref Seq ID	Gene Symbol	P-value	Meth Diff	Mean Cluster A	Mean Cluster B	CpG Cluster	CpG Island
MSPI0406S00450835	chr8	NM_030974	SHARPIN	0.0000	1.72	0.00	-1.72	YES	-
MSPI0406S00451398	chr8	NM_016208	VPS28	0.0007	1.67	2.18	0.51	YES	-
MSPI0406S00459565	chr9	NM_003995	NPR2	0.0000	1.56	-0.32	-1.88	YES	-
MSPI0406S00478974	chr9	NM_138424	KIF12	0.0002	1.70	1.74	0.04	YES	YES
MSPI0406S00481107	chr9	BF513822	DAB2IP	0.0000	1.51	1.93	0.42	-	-
MSPI0406S00486447	chr9	NM_004003, NM_021131	CRAT, PPP2R4	0.0000	1.86	0.41	-1.45	YES	-
MSPI0406S00489085	chr9	AB011087	KIAA0515	0.0000	1.90	1.56	-0.33	-	-
MSPI0406S00494431	chr9	NA		0.0001	1.58	-1.35	-2.93	-	-
MSPI0406S00496573	chr9	NA		0.0000	1.55	1.03	-0.52	YES	YES
MSPI0406S00497723	chr10	NM_014974	DIP2C	0.0002	1.76	0.79	-0.97	YES	-
MSPI0406S00509727	chr10	AL553774	KIAA1462	0.0001	1.78	3.11	1.34	-	-
MSPI0406S00509728	chr10	AL553774	KIAA1462	0.0001	1.55	0.17	-1.38	-	-
MSPI0406S00522801	chr10	NM_173540	FUT11	0.0001	1.88	1.20	-0.68	YES	YES
MSPI0406S00523480	chr10	NM_144589	COMTD1	0.0001	1.61	0.36	-1.25	YES	YES
MSPI0406S00531525	chr10	AF267854	SLC25A28	0.0001	1.58	1.39	-0.19	-	-
MSPI0406S00531531	chr10	NM_031212	SLC25A28	0.0001	1.67	3.10	1.44	YES	YES
MSPI0406S00531671	chr10	AB023227	DNMBP	0.0002	1.75	-0.44	-2.19	-	-
MSPI0406S00532654	chr10	NM_015448, NM_013274	POLL, RP11-529I10.4	0.0000	1.61	0.50	-1.11	-	-
MSPI0406S00539049	chr10	NM_001005339	RGS10	0.0006	1.88	2.54	0.66	YES	YES
MSPI0406S00550614	chr11	BC002550	H19	0.0001	1.51	1.00	-0.50	YES	YES
MSPI0406S00577268	chr11	NM_005169	PHOX2A	0.0003	1.74	1.45	-0.30	-	-
MSPI0406S00577800	chr11	NM_002564	P2RY2	0.0001	1.89	2.20	0.31	YES	YES
MSPI0406S00585215	chr11	NM_004398	LOC401533	0.0000	1.83	0.98	-0.85	YES	YES
MSPI0406S00592025	chr11	NM_014312	VSIG2	0.0000	1.65	1.74	0.09	YES	-
MSPI0406S00600155	chr12	NM_001769	CD9	0.0000	1.75	2.13	0.39	YES	YES
MSPI0406S00600647	chr12	NM_016162	ING4	0.0002	1.66	1.57	-0.09	-	-
MSPI0406S00600942	chr12	Z22814	ATN1	0.0000	2.13	-0.11	-2.24	-	-
MSPI0406S00601262	chr12	NM_020634	GDF3	0.0000	1.98	1.43	-0.55	-	-
MSPI0406S00601366	chr12	NM_024865	NANOG	0.0006	1.71	-0.51	-2.22	-	-
MSPI0406S00604787	chr12	NM_153207	AEBP2	0.0001	1.55	-0.34	-1.89	YES	-
MSPI0406S00605562	chr12	NM_144667	FLJ32894	0.0002	1.64	-1.18	-2.82	-	-
MSPI0406S00611736	chr12	NM_147190	LASS5	0.0001	1.65	0.69	-0.95	YES	-
MSPI0406S00614653	chr12	NM_020370	GPR84	0.0008	2.10	1.79	-0.31	-	-
MSPI0406S00616342	chr12	NM_004990	MARS	0.0000	1.96	0.68	-1.29	YES	-
MSPI0406S00619875	chr12	NM_024685	BBS10	0.0000	1.59	0.98	-0.61	YES	-
MSPI0406S00620956	chr12	NM_005447	PAMCI	0.0005	1.97	2.10	0.13	-	-
MSPI0406S00621954	chr12	NM_199040	NUDT4P1	0.0001	2.50	2.02	-0.48	YES	YES
MSPI0406S00629691	chr12	NM_017901	TPCN1	0.0004	1.54	0.85	-0.68	-	-
MSPI0406S00633671	chr12	NM_004276	CABP1	0.0005	1.62	0.99	-0.63	-	-
MSPI0406S00663882	chr13	NA		0.0000	1.57	-1.11	-2.68	-	YES
MSPI0406S00679968	chr14	AI431597	WDR22	0.0000	1.58	-1.06	-2.64	YES	-
MSPI0406S00697563	chr14	AK098354	PACS2	0.0000	2.14	1.13	-1.01	-	-
MSPI0406S00705935	chr15	NM_001211	BUB1B	0.0000	1.84	1.87	0.03	YES	YES
MSPI0406S00706182	chr15	NM_014952	BAHD1	0.0000	1.50	2.40	0.89	-	-

HELP probe sets	**Chr.**	**Ref Seq ID**	**Gene Symbol**	**P-value**	**Meth Diff**	**Mean Cluster A**	**Mean Cluster B**	**CpG Cluster**	**CpG Island**
MSPI0406S00708457	chr15	NM_025165	ELL3	0.0000	1.74	2.74	1.01	YES	YES
MSPI0406S00716590	chr15	NM_005902	SMAD3	0.0003	1.56	3.05	1.49	YES	-
MSPI0406S00718319	chr15	NM_018003	UACA	0.0007	1.73	1.34	-0.39	YES	YES
MSPI0406S00720842	chr15	NM_001042581	SNUPN	0.0000	1.99	2.06	0.08	YES	-
MSPI0406S00723766	chr15	NM_032246	MEX3B	0.0001	1.58	2.23	0.65	YES	YES
MSPI0406S00730480	chr15	NM_183376	ARRDC4	0.0002	1.62	2.48	0.86	YES	YES
MSPI0406S00731394	chr15	NM_152449	LYSMD4	0.0003	1.55	-0.22	-1.77	YES	YES
MSPI0406S00735266	chr16	NM_001037125	UNKL	0.0000	1.50	1.42	-0.09	YES	-
MSPI0406S00735606	chr16	AB037847	CRAMP1L	0.0000	1.94	0.68	-1.27	-	-
MSPI0406S00736080	chr16	NM_001099456	NPW	0.0004	1.53	1.00	-0.53	YES	-
MSPI0406S00743447	chr16	NM_002094	GSPT1	0.0000	1.55	1.70	0.16	YES	-
MSPI0406S00755023	chr16	AB002307	SRCAP	0.0003	1.97	-1.52	-3.48	-	-
MSPI0406S00757893	chr16	BF436315	N4BP1	0.0001	2.10	1.94	-0.16	-	-
MSPI0406S00761227	chr16	NA		0.0002	1.53	1.06	-0.47	-	-
MSPI0406S00765085	chr16	NM_020457	THAP11	0.0000	1.76	1.30	-0.46	YES	-
MSPI0406S00773420	chr16	NM_001537	HSBP1	0.0002	1.69	1.60	-0.09	-	-
MSPI0406S00773421	chr16	NM_001537	HSBP1	0.0000	2.11	1.69	-0.42	YES	YES
MSPI0406S00777575	chr16	AA758799	KLHDC4	0.0000	1.74	-0.06	-1.80	YES	-
MSPI0406S00780667	chr16	NM_145039	MGC16385	0.0001	1.68	0.85	-0.83	YES	-
MSPI0406S00813610	chr17	NM_000088	COL1A1	0.0004	1.50	2.57	1.07	-	-
MSPI0406S00821398	chr17	NM_002816	PSMD12	0.0002	1.77	1.03	-0.74	YES	YES
MSPI0406S00826551	chr17	NM_001258	CDK3	0.0000	1.53	-0.73	-2.26	YES	-
MSPI0406S00832779	chr17	BC020925	MGC15523	0.0008	1.73	-0.39	-2.12	-	-
MSPI0406S00834302	chr17	NM_173620, NM_175902	FLJ22222, HEXDC	0.0001	1.69	1.30	-0.39	YES	-
MSPI0406S00856224	chr18	BE676488	MBP	0.0004	1.84	-0.07	-1.92	-	-
MSPI0406S00856242	chr18	NA		0.0010	1.69	0.30	-1.39	-	-
MSPI0406S00865599	chr19	NM_015015	JMJD2B	0.0006	1.64	2.47	0.83	YES	YES
MSPI0406S00898635	chr19	AI275597	GRLF1	0.0000	1.91	0.54	-1.38	-	-
MSPI0406S00900721	chr19	NM_182575	IZUMO1	0.0008	1.55	2.41	0.86	-	-
MSPI0406S00906993	chr19	NM_017607	PPP1R12C	0.0000	1.87	1.86	-0.01	YES	-
MSPI0406S00920699	chr20	NA		0.0002	1.86	0.03	-1.83	-	-
MSPI0406S00943034	chr21	NM_001186	BACH1	0.0000	1.80	0.69	-1.11	YES	YES
MSPI0406S00957389	chr22	NM_153334	SCARF2	0.0001	1.62	1.11	-0.51	-	-
MSPI0406S00959645	chr22	NA		0.0000	1.89	0.14	-1.75	-	-
MSPI0406S00967178	chr22	NM_001003681	HMG2L1	0.0003	1.57	-1.02	-2.59	YES	YES
MSPI0406S00973723	chr22	NM_033318	C22orf32	0.0000	1.71	1.86	0.15	-	-
MSPI0406S00990002	chrX	NM_031892	SH3KBP1	0.0000	1.95	2.33	0.38	YES	-

Figure S1. Validation of HELP methylation levels by MassARRAY Epityper
Dot plot showing correlation between $\log_2$ (HpaII/MspI) ratios (x-axis) and percentage of methylation as determined by MassARRAY Epityper (y-axis) for 15 probe sets in 13 randomly selected *EVI1* AML patients. The correlation coefficient is shown.

Figure S2. *EVI1* AML patients have a distinct genome-wide methylation profile compared to other recurrent molecular and cytogenetic AML cases
The correlation plot shows the Pearson correlation of the high variance probe set methylation levels between 344 AML patients. The 16 clusters that were defined by Figueroa et al[1], are labeled accordingly and the most recurrent cytogenetic or molecular abnormalities, are shown. Clusters #2 and #8, have been identified as AML subgroups with unique methylation signatures, without common genetic or cytogenetic abnormalities and contain the majority of *EVI1+* patients (labeled red for *EVI1+* and green for *EVI1-*).

Figure S3. EVI1 binds hypermethylated genes containing EVI1 binding sequences
Quantitative PCR of chromatin immunoprecipitation in the *EVI1* positive (*EVI1+*) SB1960CB cell line (A) and the K562 cell line (B) and the *EVI1* negative (*EVI1-*) MOLM13 cell line using EVI1 and IgG antibody. Percentage of amount of input material is shown. The mean of two independent experiments is shown. The EVI1 binding sequence present in the promoter region of the hypermethylated genes is depected.

Figure S4. *DNMT3A* overexpressed in *EVI1* AMLs
A gene expression correlation view of 285 AMLs as previously described[2] shows the expression of *DNMT3A* in bars per patient. Focusing on the clusters highly over-represented with *EVI1* AMLs a trend for *DNTM3A* overexpression is seen.

Figure S5. Unsupervised hierarchical clustering using genome wide methylation levels in *EVI1* AMLs revealed two main clusters
Unsupervised cluster analysis using Pearson's correlation with Ward's method revealed two distinct *EVI1* subclusters; A and B. Using 10.000 bootstraps the Approximately Unbiased (AU) and Bootstrap Probability (BP) percentages were calculated per tree in the hierarchical clustering to show the tightness per subcluster, i.e. higher percentage means tighter cluster.

REFERENCES

1. Figueroa ME, Lugthart S, Li Y, et al. DNA Methylation Signatures Identify Biologically Distinct Subtypes in Acute Myeloid Leukemia. *Cancer Cell*.
2. Valk PJ, Verhaak RG, Beijen MA, et al. Prognostically useful gene-expression profiles in acute myeloid leukemia. *N Engl J Med*. 2004;350:1617–1628.

Figure S1

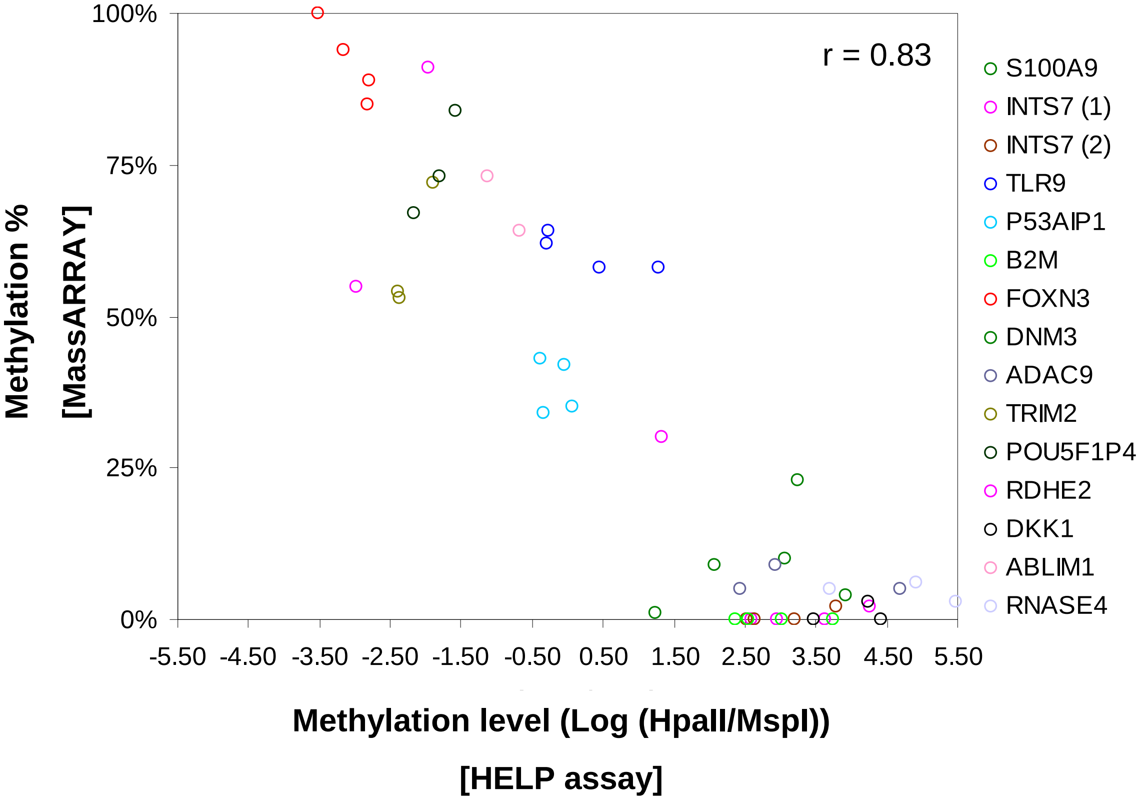

r = 0.83
Methylation %
[MassARRAY]
100%
75%
50%
25%
0%
-5.50
-4.50
-3.50
-2.50
-1.50
-0.50
0.50
1.50
2.50
3.50
4.50
5.50
Methylation level (Log (HpaII/MspI))
[HELP assay]
S100A9
INTS7 (1)
INTS7 (2)
TLR9
P53AIP1
B2M
FOXN3
DNM3
ADAC9
TRIM2
POU5F1P4
RDHE2
DKK1
ABLIM1
RNASE4

Figure S2

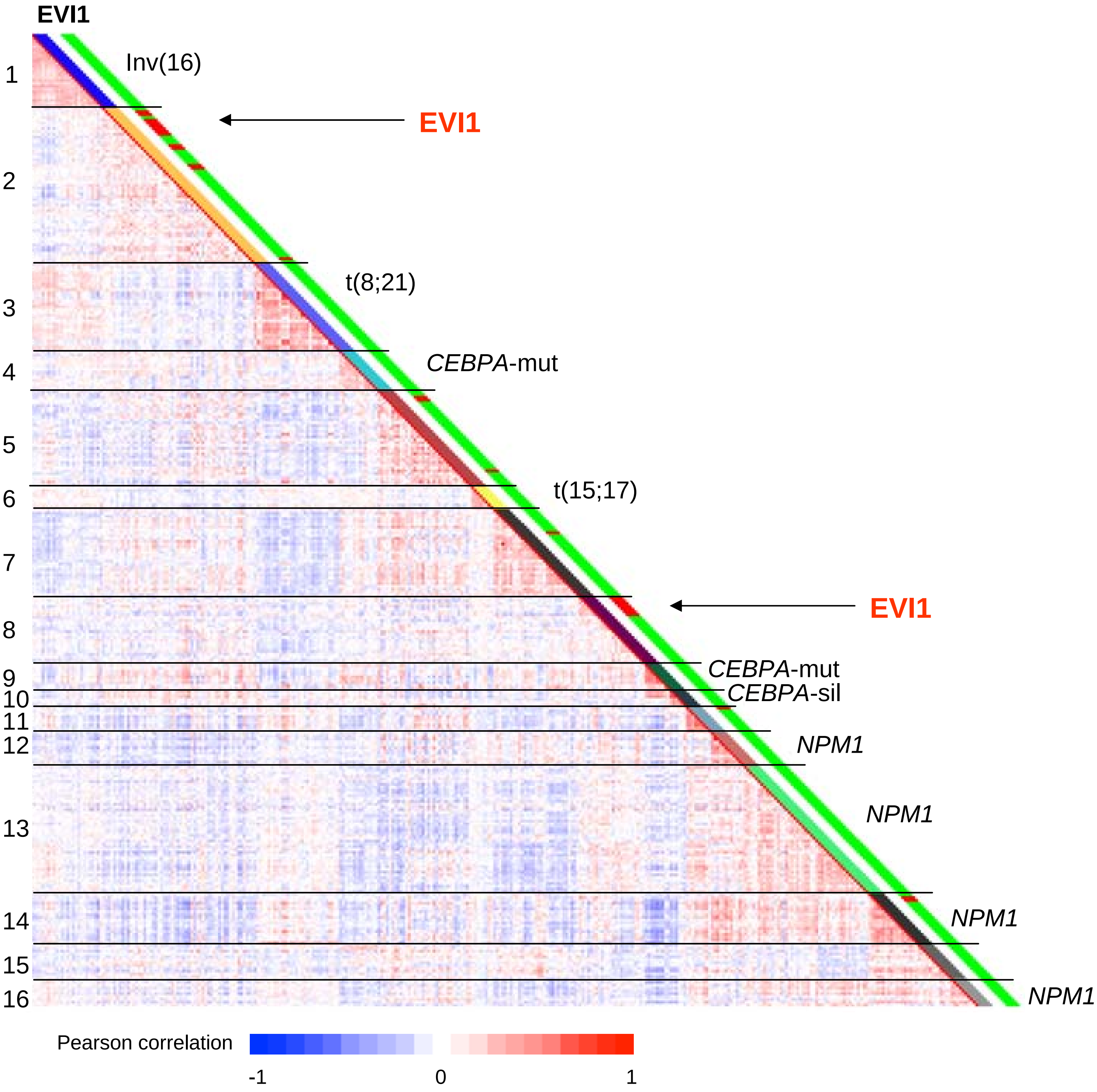

EVI1
Inv(16)
EVI1
t(8;21)
CEBPA-mut
t(15;17)
EVI1
CEBPA-mut
CEBPA-sil
NPM1
NPM1
NPM1
NPM1
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
Pearson correlation
-1
0
1

Figure S3

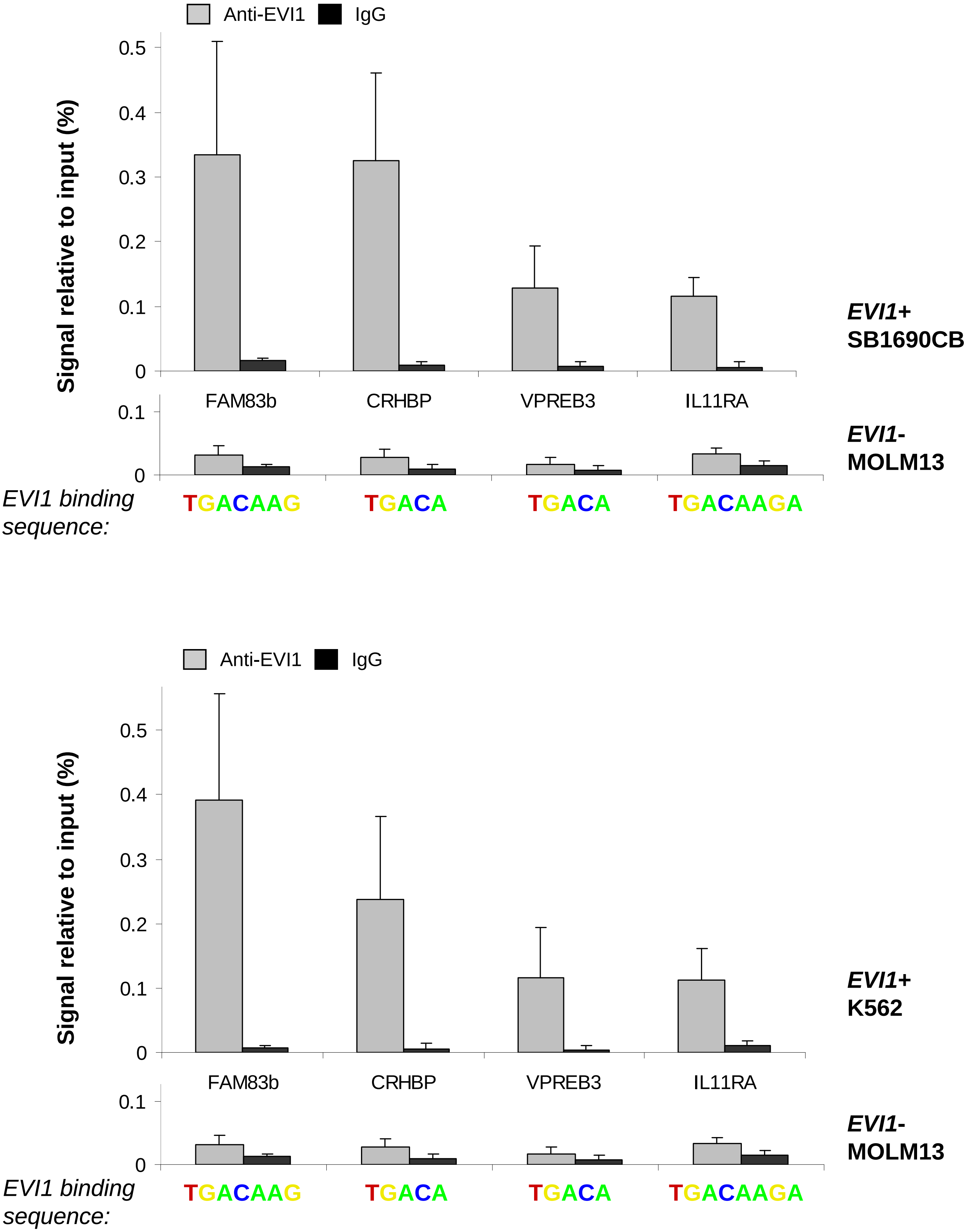

Anti-EVI1
IgG
Signal relative to input (%)
0.5
0.4
0.3
0.2
0.1
0
FAM83b
CRHBP
VPREB3
IL11RA
EVI1+
SB1690CB
0.1
0
EVI1-
MOLM13
EVI1 binding sequence:
TGACAAG
TGACA
TGACA
TGACAAGA
Anti-EVI1
IgG
Signal relative to input (%)
0.5
0.4
0.3
0.2
0.1
0
FAM83b
CRHBP
VPREB3
IL11RA
EVI1+
K562
0.1
0
EVI1-
MOLM13
EVI1 binding sequence:
TGACAAG
TGACA
TGACA
TGACAAGA

Figure S4

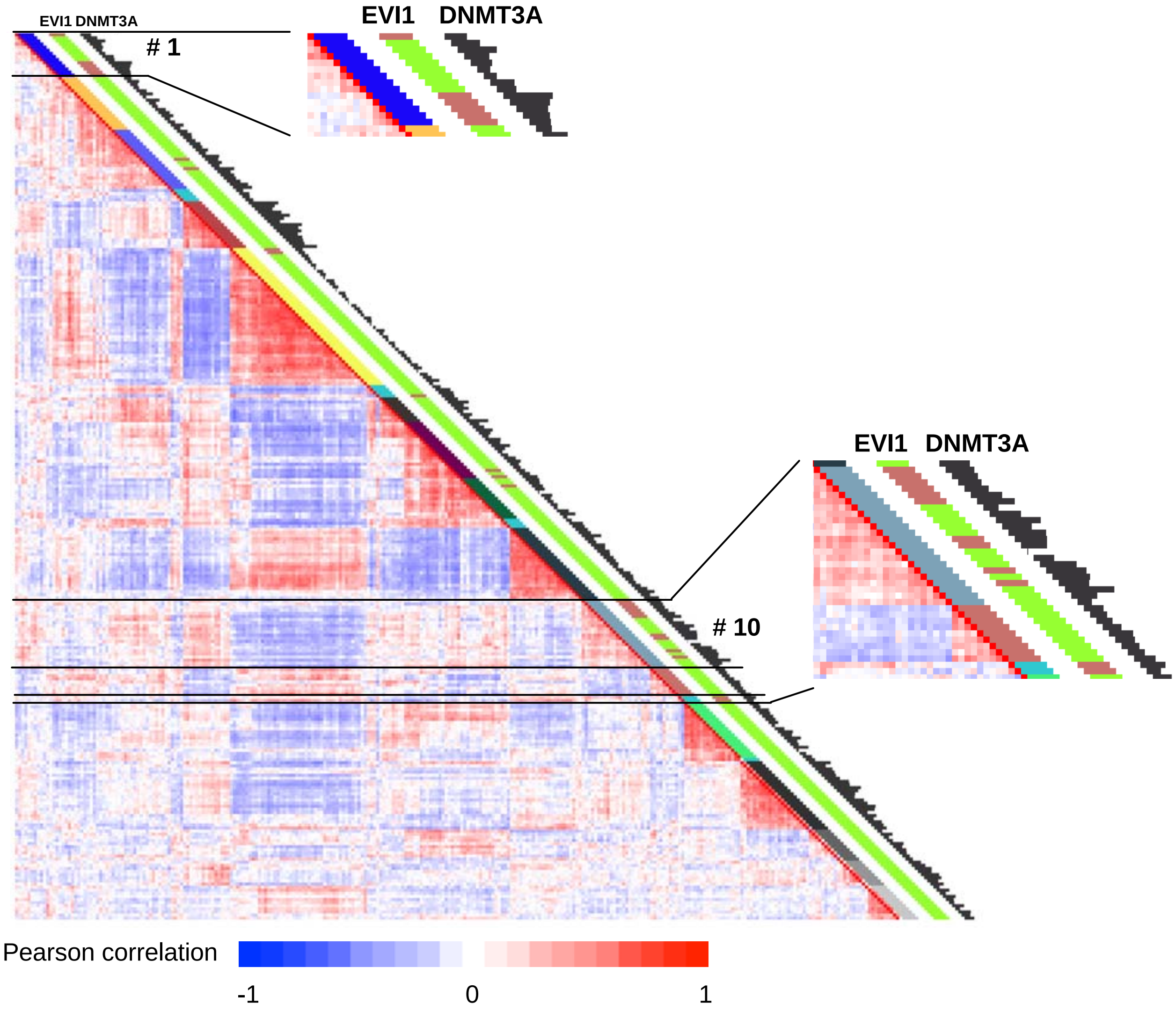
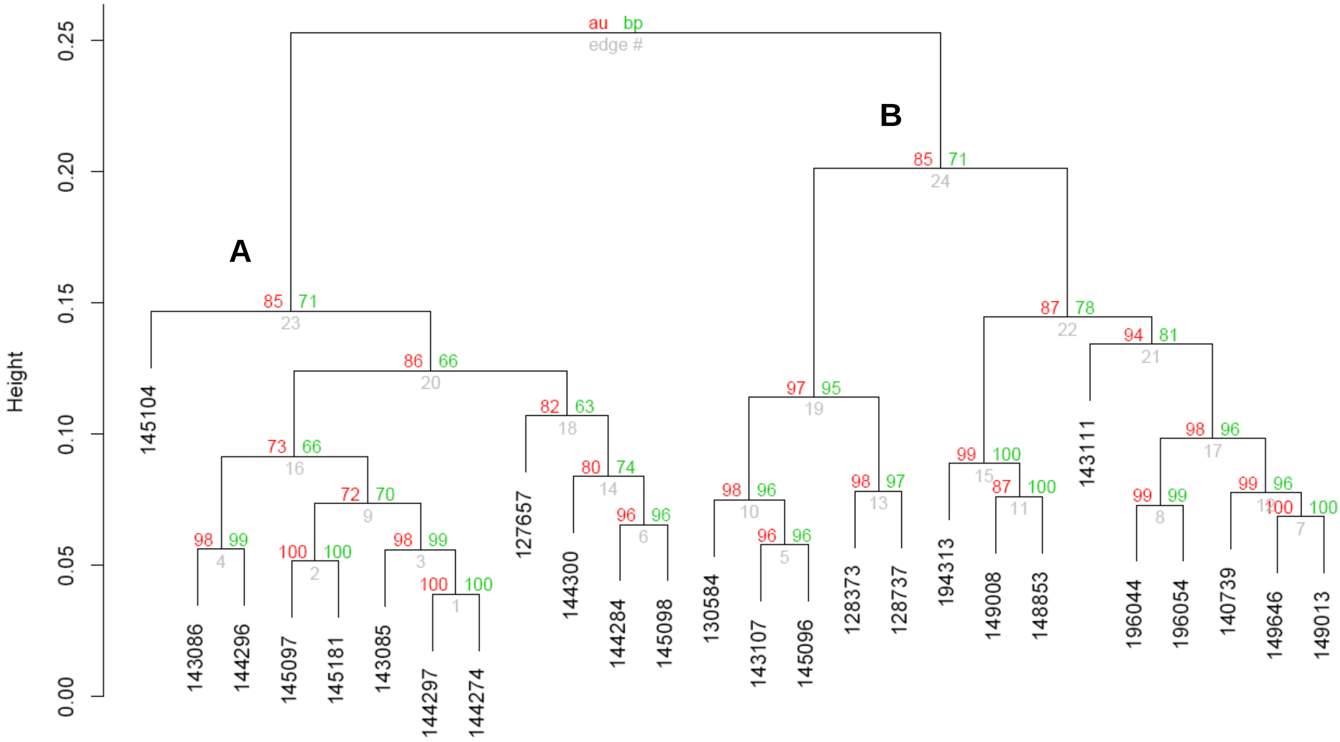

EVI1 DNMT3A
1
EVI1
DNMT3A
EVI1
DNMT3A
10
Pearson correlation
-1
0
1

Figure S5

Pearson’s Correlation distance and Ward’s Method



10.000 Bootstraps
p-value -Approximately Unbiased
-Bootstrap Probability
} > 95% = significant