

Interferon α -induced SAMHD1 regulates human cultured megakaryocyte apoptosis and proplatelet formation

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Supplemental Data

Interferon Alpha-Induced SAMHD1 Regulates Human Cultured Megakaryocyte
Apoptosis and Proplatelet Formation

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Supplemental Table 1. Changes in mRNA levels in differentiating human CD61+ cultured MKs on INF α stimulation.

EnsemblID	geneName	CONTROL- treated baseMean	INF α - TREATED baseMean	log2 fold change	p-value	adjusted p-value
				INF α CONTROL		
ENSG00000101347.9	SAMHD1	557.4	9633.0	4.11	6.74E-23	2.02E-18
ENSG00000136147.18	PHF11	243.9	1393.7	2.52	1.88E-22	2.82E-18
ENSG00000110057.8	UNC93B1	194.5	1310.9	2.75	1.19E-21	1.19E-17
ENSG00000172183.15	ISG20	177.7	3460.1	4.28	1.43E-15	1.07E-11
ENSG00000206190.11	ATP10A	24.6	520.3	4.41	1.85E-15	1.11E-11
ENSG00000142089.16	IFITM3	1237.0	7117.0	2.52	3.01E-15	1.51E-11
ENSG00000025708.14	TYMP	927.5	3560.7	1.94	1.21E-14	5.20E-11
ENSG00000185885.16	IFITM1	569.3	8065.2	3.82	3.22E-12	1.21E-08
ENSG00000110446.11	SLC15A3	13.0	215.4	4.05	7.15E-11	2.14E-07
ENSG00000138035.15	PNPT1	2179.2	7841.0	1.85	7.89E-11	2.15E-07
ENSG00000106785.15	TRIM14	148.3	1284.1	3.11	1.68E-10	4.20E-07
ENSG00000174808.12	BTC	33.6	221.8	2.72	2.95E-10	6.81E-07
ENSG00000130303.13	BST2	26.2	393.1	3.91	4.04E-10	8.65E-07
ENSG00000168394.11	TAP1	3302.4	12296.9	1.90	5.31E-10	9.95E-07
ENSG00000156587.16	UBE2L6	2131.2	11836.3	2.47	6.84E-10	1.21E-06
ENSG00000140511.11	HAPLN3	95.4	303.7	1.67	8.05E-10	1.31E-06
ENSG00000204642.14	HLA-F	803.2	2380.4	1.57	8.33E-10	1.31E-06
ENSG00000223960.6	PRKRA-AS1	172.1	608.8	1.82	1.46E-09	2.16E-06
ENSG00000240065.8	PSMB9	233.0	1127.3	2.27	1.52E-09	2.16E-06
ENSG00000178685.14	PARP10	1483.9	7248.1	2.29	4.87E-09	6.64E-06
ENSG00000135114.12	OASL	175.8	2279.7	3.70	1.42E-08	1.85E-05
ENSG00000125148.7	MT2A	96.3	758.6	2.98	1.55E-08	1.87E-05
ENSG00000116663.11	FBXO6	49.7	370.6	2.90	1.56E-08	1.87E-05
ENSG00000169248.12	CXCL11	528.5	2782.7	2.40	2.16E-08	2.43E-05
ENSG00000204267.14	TAP2	1314.2	5839.7	2.15	2.20E-08	2.43E-05
ENSG00000204264.10	PSMB8	2282.2	6010.9	1.40	2.27E-08	2.43E-05
ENSG00000108771.13	DHX58	730.8	5133.4	2.81	4.98E-08	4.98E-05
ENSG00000068079.7	IFI35	469.3	4305.6	3.20	5.64E-08	5.45E-05
ENSG00000173821.19	AC124319.1	9315.4	45655.9	2.29	7.29E-08	6.83E-05
ENSG00000221963.6	APOL6	1240.5	9540.8	2.94	1.03E-07	9.38E-05
ENSG00000002549.12	LAP3	5182.0	17514.4	1.76	2.13E-07	1.88E-04
ENSG00000137200.13	CMTR1	1800.0	5606.7	1.64	2.30E-07	1.97E-04
ENSG00000196141.14	SPATS2L	204.1	1161.9	2.51	2.61E-07	2.17E-04

ENSG00000198133.8	TMEM229B	112.2	348.5	1.63	2.74E-07	2.22E-04
ENSG00000140807.7	NKD1	168.1	596.2	1.83	2.86E-07	2.26E-04
ENSG00000185201.16	IFITM2	693.9	1877.8	1.44	3.16E-07	2.43E-04
ENSG00000130813.18	C19orf66	1155.3	4420.1	1.94	4.75E-07	3.56E-04
ENSG00000133943.20	DGLUCY	916.7	2003.2	1.13	7.73E-07	5.65E-04
ENSG00000184979.9	USP18	2711.5	13730.4	2.34	1.11E-06	7.89E-04
ENSG00000168961.17	LGALS9	92.9	453.3	2.29	1.26E-06	8.57E-04
ENSG00000163131.11	CTSS	129.1	422.1	1.71	1.47E-06	9.55E-04
ENSG00000181381.13	DDX60L	3513.3	24126.3	2.78	1.50E-06	9.59E-04
ENSG00000157601.14	MX1	5023.1	83193.7	4.05	2.62E-06	1.54E-03
ENSG00000188313.13	PLSCR1	3800.9	15297.0	2.01	2.62E-06	1.54E-03
ENSG00000173786.17	CNP	2354.2	6531.4	1.47	3.79E-06	2.18E-03
ENSG00000149131.15	SERPING1	87.5	316.1	1.85	5.41E-06	3.00E-03
ENSG00000177989.13	ODF3B	39.6	200.9	2.34	7.23E-06	3.87E-03
ENSG00000133106.14	EPST11	4814.2	24961.0	2.37	8.60E-06	4.52E-03
ENSG00000125826.20	RBCK1	1438.8	3844.5	1.42	9.22E-06	4.76E-03
ENSG00000060491.16	OGFR	564.4	1158.6	1.04	9.52E-06	4.84E-03
ENSG00000130589.16	HELZ2	1740.5	9537.4	2.45	1.21E-05	6.02E-03
ENSG00000111331.13	OAS3	2113.8	24324.4	3.52	1.30E-05	6.39E-03
ENSG00000138760.10	SCARB2	457.4	2349.6	2.36	1.41E-05	6.83E-03
ENSG00000132109.10	TRIM21	1617.5	5655.1	1.81	1.45E-05	6.90E-03
ENSG00000013364.19	MVP	870.1	1807.7	1.05	1.60E-05	7.48E-03
ENSG00000165806.19	CASP7	525.9	1444.6	1.46	1.88E-05	8.66E-03
ENSG00000010030.14	ETV7	204.6	527.5	1.37	1.93E-05	8.77E-03
ENSG00000035720.8	STAP1	37.7	180.1	2.26	1.99E-05	8.91E-03
ENSG00000177409.12	SAMD9L	1886.0	15436.3	3.03	2.38E-05	1.05E-02
ENSG00000239713.9	APOBEC3G	1232.6	3026.7	1.30	3.13E-05	1.36E-02
ENSG00000100342.20	APOL1	44.5	469.4	3.40	4.49E-05	1.90E-02
ENSG00000038210.13	PI4K2B	1754.2	3423.9	0.96	6.54E-05	2.58E-02
ENSG00000170581.14	STAT2	3168.3	10928.3	1.79	6.84E-05	2.66E-02
ENSG00000086065.14	CHMP5	3603.8	7951.9	1.14	8.57E-05	3.25E-02
ENSG00000160932.11	LY6E	2512.2	12768.5	2.35	9.51E-05	3.56E-02
ENSG00000134326.11	CMPK2	4480.2	32352.2	2.85	1.29E-04	4.72E-02
ENSG00000137628.17	DDX60	2923.1	14443.5	2.30	1.46E-04	5.25E-02
ENSG00000115415.18	STAT1	12649.1	50923.9	2.01	1.48E-04	5.27E-02
ENSG00000188157.15	AGRN	205.7	724.4	1.82	1.57E-04	5.40E-02
ENSG00000256043.3	CTSO	107.3	279.4	1.38	1.78E-04	6.08E-02
ENSG00000185507.20	IRF7	1450.1	7474.6	2.37	1.92E-04	6.48E-02
ENSG00000166750.10	SLFN5	140.7	811.0	2.53	2.04E-04	6.79E-02
ENSG00000059378.12	PARP12	2629.1	8620.5	1.71	2.26E-04	7.46E-02

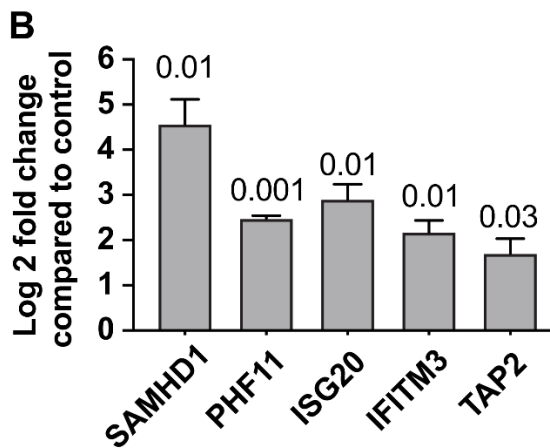
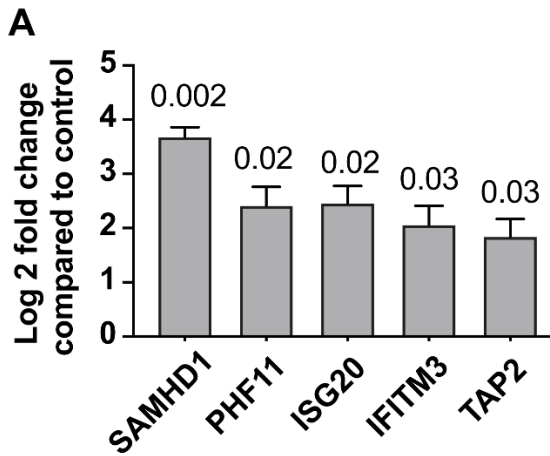
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ENSG00000111335.12	OAS2	839.3	16867.7	4.33	2.63E-04	8.40E-02
ENSG00000155363.18	MOV10	1808.9	6272.3	1.79	3.22E-04	1.02E-01
ENSG00000128335.14	APOL2	248.5	1142.5	2.20	3.43E-04	1.07E-01
ENSG00000196684.12	HSH2D	46.4	204.2	2.14	3.85E-04	1.18E-01
ENSG00000169245.6	CXCL10	77.1	1694.9	4.46	4.50E-04	1.35E-01
ENSG00000183486.13	MX2	880.3	15233.7	4.11	4.71E-04	1.40E-01
ENSG00000196116.8	TDRD7	978.1	2755.3	1.49	4.99E-04	1.45E-01
ENSG00000132274.16	TRIM22	785.6	13614.9	4.12	5.06E-04	1.46E-01
ENSG00000185404.16	SP140L	227.1	454.5	1.00	5.53E-04	1.58E-01
ENSG00000182240.16	BACE2	326.3	624.1	0.94	5.99E-04	1.69E-01
ENSG00000204525.16	HLA-C	6849.8	11846.0	0.79	6.75E-04	1.87E-01
ENSG00000108679.13	LGALS3BP	1011.0	3343.3	1.73	7.43E-04	2.02E-01
ENSG00000169228.14	RAB24	619.0	1071.7	0.79	7.73E-04	2.07E-01
ENSG00000105402.8	NAPA	3170.3	6024.1	0.93	8.40E-04	2.23E-01
ENSG00000164054.15	SHISA5	4357.2	11322.2	1.38	9.29E-04	2.44E-01
ENSG00000136514.3	RTP4	198.6	1332.3	2.75	9.58E-04	2.50E-01
ENSG00000138642.14	HERC6	2667.8	14155.6	2.41	9.97E-04	2.56E-01
ENSG00000169660.16	HEXD	433.7	839.0	0.95	1.05E-03	2.68E-01
ENSG00000183831.6	ANKRD45	441.0	1861.4	2.08	1.09E-03	2.73E-01
ENSG00000104419.14	NDRG1	533.8	944.9	0.82	1.09E-03	2.73E-01
ENSG00000166801.15	FAM111A	899.4	1768.6	0.98	1.26E-03	3.07E-01
ENSG00000172667.11	ZMAT3	4623.1	2197.7	-1.07	1.34E-03	3.21E-01
ENSG00000089127.13	OAS1	198.7	3332.0	4.07	1.36E-03	3.21E-01
ENSG00000128284.19	APOL3	202.3	664.1	1.72	1.40E-03	3.29E-01
ENSG00000132530.17	XAF1	1251.3	7526.3	2.59	1.66E-03	3.83E-01
ENSG00000275023.4	MLLT6	1237.6	1963.0	0.67	1.68E-03	3.84E-01
ENSG00000106560.11	GIMAP2	209.8	386.9	0.88	1.70E-03	3.86E-01
ENSG00000115271.11	GCA	905.5	1606.0	0.83	1.87E-03	4.10E-01
ENSG00000067066.17	SP100	2915.1	8358.1	1.52	1.94E-03	4.22E-01
ENSG00000134627.12	PIWIL4	157.3	306.8	0.96	1.99E-03	4.29E-01
ENSG00000013374.16	NUB1	1242.8	2370.0	0.93	2.03E-03	4.34E-01
ENSG00000137965.11	IFI44	1752.1	10207.0	2.54	2.09E-03	4.44E-01
ENSG00000168016.14	TRANK1	2174.0	4288.8	0.98	2.12E-03	4.47E-01
ENSG00000234883.5	MIR155HG	337.6	1267.9	1.91	2.15E-03	4.49E-01
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ENSG00000121858.11	TNFSF10	31.0	733.3	4.56	2.57E-03	5.23E-01
ENSG00000185880.13	TRIM69	497.1	1365.4	1.46	2.59E-03	5.25E-01
ENSG00000160710.16	ADAR	16087.8	39436.4	1.29	2.80E-03	5.60E-01

ENSG00000107201.10	DDX58	3634.6	19534.0	2.43	3.12E-03	6.01E-01
ENSG00000133313.15	CNDP2	1520.8	2520.5	0.73	3.16E-03	6.02E-01
ENSG00000135899.17	SP110	532.7	2121.5	1.99	3.17E-03	6.02E-01
ENSG00000138496.16	PARP9	5077.8	18355.8	1.85	3.24E-03	6.11E-01
ENSG00000114450.10	GNB4	5897.1	10114.2	0.78	3.27E-03	6.13E-01
ENSG00000163121.10	NEURL3	82.3	255.2	1.63	3.54E-03	6.52E-01
ENSG00000111801.16	BTN3A3	65.1	141.9	1.12	3.79E-03	6.84E-01
ENSG00000175550.8	DRAP1	2880.9	6584.7	1.19	3.90E-03	6.99E-01
ENSG00000123609.10	NMI	1461.8	3073.6	1.07	4.21E-03	7.51E-01
ENSG00000205268.10	PDE7A	1835.2	3504.6	0.93	4.23E-03	7.51E-01
ENSG00000140464.19	PML	492.4	1368.9	1.48	4.28E-03	7.54E-01
ENSG00000168404.13	MLKL	448.8	923.3	1.04	4.39E-03	7.70E-01
ENSG00000258659.6	TRIM34	301.8	1200.3	1.99	4.77E-03	8.26E-01
ENSG00000132256.18	TRIM5	2165.4	4533.9	1.07	5.03E-03	8.62E-01
ENSG00000205413.8	SAMD9	9093.0	26651.4	1.55	5.28E-03	8.96E-01
ENSG00000172936.14	MYD88	346.7	730.7	1.08	5.32E-03	8.96E-01
ENSG00000205220.12	PSMB10	1328.1	2172.8	0.71	5.37E-03	8.99E-01
ENSG00000076321.11	KLHL20	2353.8	4469.1	0.93	6.06E-03	9.93E-01
ENSG00000100911.16	PSME2	1510.9	2628.9	0.80	6.29E-03	1.00E+00
ENSG00000204397.7	CARD16	135.4	276.5	1.03	6.44E-03	1.00E+00
ENSG00000092010.15	PSME1	2756.4	4333.7	0.65	6.47E-03	1.00E+00
ENSG00000163840.10	DTX3L	7989.0	24118.3	1.59	6.67E-03	1.00E+00
ENSG00000115267.7	IFIH1	1604.1	5665.2	1.82	6.92E-03	1.00E+00
ENSG00000127951.7	FGL2	162.2	365.2	1.17	7.10E-03	1.00E+00
ENSG00000112773.16	TENT5A	1159.0	2178.7	0.91	7.49E-03	1.00E+00
ENSG00000234127.9	TRIM26	132.7	311.1	1.23	7.57E-03	1.00E+00
ENSG00000105835.12	NAMPT	2757.5	6061.2	1.14	7.64E-03	1.00E+00
ENSG00000126709.15	IFI6	5259.8	24629.7	2.23	8.03E-03	1.00E+00
ENSG00000100226.16	GTPBP1	1432.1	2591.6	0.86	8.05E-03	1.00E+00
ENSG00000263740.2	RN7SL4P	5260.4	8890.4	0.76	8.17E-03	1.00E+00
ENSG00000164136.17	IL15	97.9	186.0	0.93	8.32E-03	1.00E+00
ENSG00000163644.15	PPM1K	3908.4	7376.0	0.92	8.64E-03	1.00E+00
ENSG00000197536.11	C5orf56	116.7	476.8	2.03	8.69E-03	1.00E+00
ENSG00000184992.11	BRI3BP	1098.7	600.4	-0.87	9.29E-03	1.00E+00
ENSG00000135052.16	GOLM1	392.6	898.0	1.19	9.64E-03	1.00E+00
ENSG00000106034.18	CPED1	901.5	2310.6	1.36	1.07E-02	1.00E+00
ENSG00000134321.12	RSAD2	7453.0	87283.2	3.55	1.07E-02	1.00E+00
ENSG00000165949.12	IFI27	94.8	1172.9	3.63	1.10E-02	1.00E+00
ENSG00000151338.18	MIPOL1	82.9	141.4	0.77	1.10E-02	1.00E+00
ENSG00000213689.14	TREX1	508.0	1006.3	0.99	1.13E-02	1.00E+00

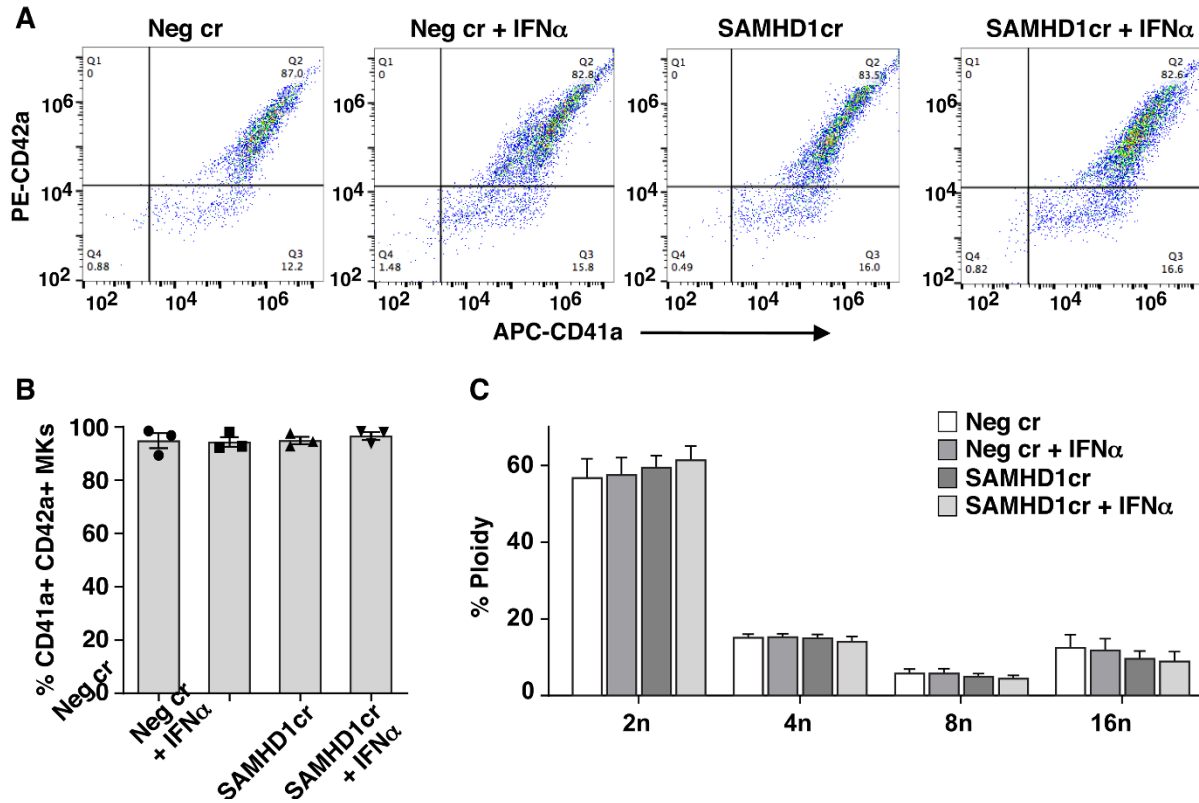
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ENSG00000140105.18	WARS	3870.2	9213.5	1.25	1.29E-02	1.00E+00
ENSG00000129226.14	CD68	4152.3	6509.5	0.65	1.46E-02	1.00E+00
ENSG00000137767.14	SQOR	2011.6	3994.9	0.99	1.51E-02	1.00E+00
ENSG00000140853.15	NLRC5	5700.0	10202.1	0.84	1.54E-02	1.00E+00
ENSG00000187608.9	ISG15	4297.0	33110.7	2.95	1.60E-02	1.00E+00
ENSG00000137496.17	IL18BP	75.6	139.0	0.88	1.64E-02	1.00E+00
ENSG00000261609.6	GAN	1823.9	1141.0	-0.68	1.80E-02	1.00E+00
ENSG00000102524.11	TNFSF13B	1954.7	7555.9	1.95	1.80E-02	1.00E+00
ENSG00000276161.1	SNORA17B	276.2	180.5	-0.61	1.83E-02	1.00E+00
ENSG00000118507.17	AKAP7	169.2	282.1	0.74	2.07E-02	1.00E+00
ENSG00000130775.16	THEMIS2	1535.7	2602.9	0.76	2.15E-02	1.00E+00
ENSG00000163512.14	AZI2	2040.0	3124.4	0.62	2.15E-02	1.00E+00
ENSG00000234745.11	HLA-B	34259.0	53980.6	0.66	2.15E-02	1.00E+00
ENSG00000054219.11	LY75	119.4	255.7	1.10	2.17E-02	1.00E+00
ENSG00000152558.15	TMEM123	17122.2	27576.1	0.69	2.17E-02	1.00E+00
ENSG00000003400.14	CASP10	379.2	984.2	1.38	2.19E-02	1.00E+00
ENSG00000122694.16	GLIPR2	475.8	771.0	0.70	2.19E-02	1.00E+00
ENSG00000138166.6	DUSP5	3823.0	5876.2	0.62	2.40E-02	1.00E+00
ENSG00000168899.5	VAMP5	95.3	177.1	0.89	2.40E-02	1.00E+00
ENSG00000183688.4	RFLNB	1409.1	869.3	-0.70	2.43E-02	1.00E+00
ENSG00000000971.15	CFH	3106.7	5238.3	0.75	2.47E-02	1.00E+00
ENSG00000213928.8	IRF9	3373.7	7408.7	1.13	2.67E-02	1.00E+00
ENSG00000245556.2	SCAMP1-AS1	176.3	284.6	0.69	2.88E-02	1.00E+00
ENSG00000171714.11	ANO5	72.5	147.3	1.02	3.01E-02	1.00E+00
ENSG00000152778.9	IFIT5	2745.9	9069.5	1.72	3.06E-02	1.00E+00
ENSG00000104518.11	GSDMD	1216.8	2039.5	0.75	3.17E-02	1.00E+00
ENSG00000055332.18	EIF2AK2	5651.8	15920.6	1.49	3.27E-02	1.00E+00
ENSG00000285446.1	Z84488.2	78.6	173.7	1.14	3.42E-02	1.00E+00
ENSG00000119673.14	ACOT2	598.9	960.3	0.68	3.42E-02	1.00E+00
ENSG00000107262.22	BAG1	3181.4	4788.0	0.59	3.55E-02	1.00E+00
ENSG00000111224.14	PARP11	487.9	777.9	0.67	3.57E-02	1.00E+00
ENSG00000229474.6	PATL2	239.5	397.8	0.73	3.62E-02	1.00E+00
ENSG00000128604.20	IRF5	86.9	184.5	1.08	3.76E-02	1.00E+00
ENSG00000172123.12	SLFN12	697.7	1073.7	0.62	3.92E-02	1.00E+00
ENSG00000128512.21	DOCK4	208.2	329.0	0.66	3.92E-02	1.00E+00
ENSG00000017483.15	SLC38A5	1061.0	2055.2	0.95	4.00E-02	1.00E+00
ENSG00000146425.11	DYNLT1	864.1	1486.0	0.78	4.00E-02	1.00E+00
ENSG00000137842.7	TMEM62	546.3	834.8	0.61	4.12E-02	1.00E+00

ENSG00000167207.13	NOD2	135.8	485.6	1.84	4.17E-02	1.00E+00
ENSG00000085831.15	TTC39A	74.2	143.7	0.95	4.23E-02	1.00E+00
ENSG00000113319.13	RASGRF2	166.9	404.5	1.28	4.39E-02	1.00E+00
ENSG00000155629.15	PIK3AP1	3388.1	7823.8	1.21	4.60E-02	1.00E+00
ENSG00000259399.1	TGIF2-RAB5IF	102.4	230.5	1.17	4.78E-02	1.00E+00
ENSG00000137959.16	IFI44L	2318.0	20566.3	3.15	4.80E-02	1.00E+00
ENSG00000222489.1	SNORA79B	266.9	171.2	-0.64	4.96E-02	1.00E+00
ENSG00000078018.19	MAP2	2330.3	5761.3	1.31	4.96E-02	1.00E+00

List of cultured human MK RNA transcripts differentially expressed in response to $\text{INF}\alpha$. FPKM>1000 was averaged from three independent, day 13 cord blood-derived CD61+ MKs. Log2 fold change of $\text{INF}\alpha$ treated MKs compared to control (PBS treated) were calculated, and presented by intensity of red color for up-regulated genes or by green color for down-regulated genes. The single noncoding RNA (MIR155) is highlighted. Table is ranked based on statistical significance p-value from highest to lowest.



Supplemental Figure 1. Validation of RNA-Seq of IFN α treatment in human cultured MKs. The validation of RNA-Seq analysis (Table 1) was performed by real time PCR using (A) technical and (B) biological replicates (CD61 purified day 13 MKs treated with 1000 U/ml IFN α or PBS, three independent cords). Most differentially expressed genes by FDR < 0.05 on IFN α stimulation from RNA-seq experiment is validated here. Log₂ fold change was plotted for IFN α stimulated CD61+ MKs, normalized to PBS control. Actin was used as a housekeeping gene. Statistical significance was determined by one sample t-test. Error bars represent mean \pm standard error of mean.



Supplemental Figure 2. IFN α -induced SAMHD1 is dispensable for human MK maturation and ploidy. CRISPR/Cas9 knock-down of *SAMHD1* (denoted as ‘SAMHD1 cr’) in CD34+ derived culture was performed at day 3. On day 9, cells are treated with 1000 U/ml IFN α or PBS. Results were compared with negative control CRISPR (denoted as ‘neg cr’). (A) Day 13 cultured MKs were assayed for MK maturation by staining the cells with APC labeled anti-CD41a and PE labeled anti-CD42a antibodies at 37°C for 10 minutes. CD41a+CD42a+ MKs were analyzed by flow cytometry. Representative flow tracing is shown. (B) The percentage of CD41a+CD42a+ MKs by flow cytometry using neg cr vs. SAMHD1 cr treated with IFN α or PBS (n=3). (C) Neg cr vs. SAMHD1 cr MKs treated with IFN α or PBS were stained with APC labeled anti-CD41a antibody and propidium iodide, and ploidy was assessed by flow cytometry (n=3). No statistically significant difference was observed for B-C. Error bars represent mean \pm standard error of mean.