

ATM gene alterations in chronic lymphocytic leukemia patients induce a distinct gene expression profile and predict disease progression

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Online Supplementary Design and Methods

RNA extraction and oligonucleotide microarray

Total RNA was extracted using the RNeasy mini procedure (Qiagen), according to the manufacturer's instructions. All samples analyzed contained at least 90% leukemic cells. HGU133 Plus 2.0 gene chips (Affymetrix, Santa Clara, CA, USA) were used to determine gene expression profiles.

Briefly, first strand cDNA was synthesized from 5 µg total RNA using T7-(dT)₂₄ primers and reverse transcribed with the Roche Applied Science Microarray cDNA Synthesis kit (Mannheim, Germany); after the second strand cDNA synthesis, the product was used in an *in vitro* transcription reaction [Roche Applied Science Microarray RNA Target Synthesis (T7) kit] to generate biotinylated complementary RNA (cRNA). Eleven micrograms of fragmented cRNA were hybridized on microarrays for 16 h and subsequently gene chips were washed, stained and scanned.

Statistical methods for microarray analysis

Oligonucleotide microarray analysis was performed with the dChip software (www.dchip.org), which uses an invariant set normalization method in which the array with median overall intensity was chosen as the baseline for normalization. Model-based expressions were computed for each array and probe set using the PM-MM model.¹

Non-specific filtering criteria for unsupervised clustering required the expression level to be higher than 300 in more than 10% of the samples and the ratio of the standard deviation (SD) to the mean expression across all samples to be between 1 and 1,000. Unsupervised clustering was performed as described by Eisen *et al.*²

Real-time quantitative polymerase chain reaction analysis

One microgram of total RNA was reverse-transcribed using the Advantage RT-for-PCR Kit (Clontech, Mountain View, CA, USA). Real-time quantitative polymerase chain reaction (Q-PCR) analysis was performed with an ABI PRISM 7500 sequence detection system and SYBR green dye (Applied Biosystems). The real-time PCR conditions were as follows: one cycle at 50°C for 2 min, one cycle at 95°C for 10 min, one cycle at 95°C for 15 s, one cycle at 60°C for 1 min, for a total of 40 cycles. For each sample, *GAPDH* C_T values were utilized for normalization purposes. For each gene, relative expression levels were computed as the difference (2^{-ΔCT}) between the target gene C_T and *GAPDH* C_T.

Primers were designed by Primer Express 1.5.1 software (Applied Biosystems). Gene symbols and primers are presented in *Online Supplementary Table S2*.

Box plots and *P* values were obtained using a tool available on the website (<http://www.physics.csbsju.edu/stats/>).

References

- Li C, Wong WH. Model-based analysis of oligonucleotide arrays: expression index computation and outlier detection. *Proc Natl Acad Sci USA*. 2001;98(1):31-6.
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ATM LCDAYIILANLDATQWKTKRKGINIPADQPITKLNKLEDDVVVPTMEIKVDHTGEYGNLVT
PI3K VIDMLQKVTIDIKSLSAEKYDVSSQVVISQLKQKLENLQNLNLP---QSFVRVPYDPG-LKA
: * : : : : . . * **:*:*: : * . . : * * :

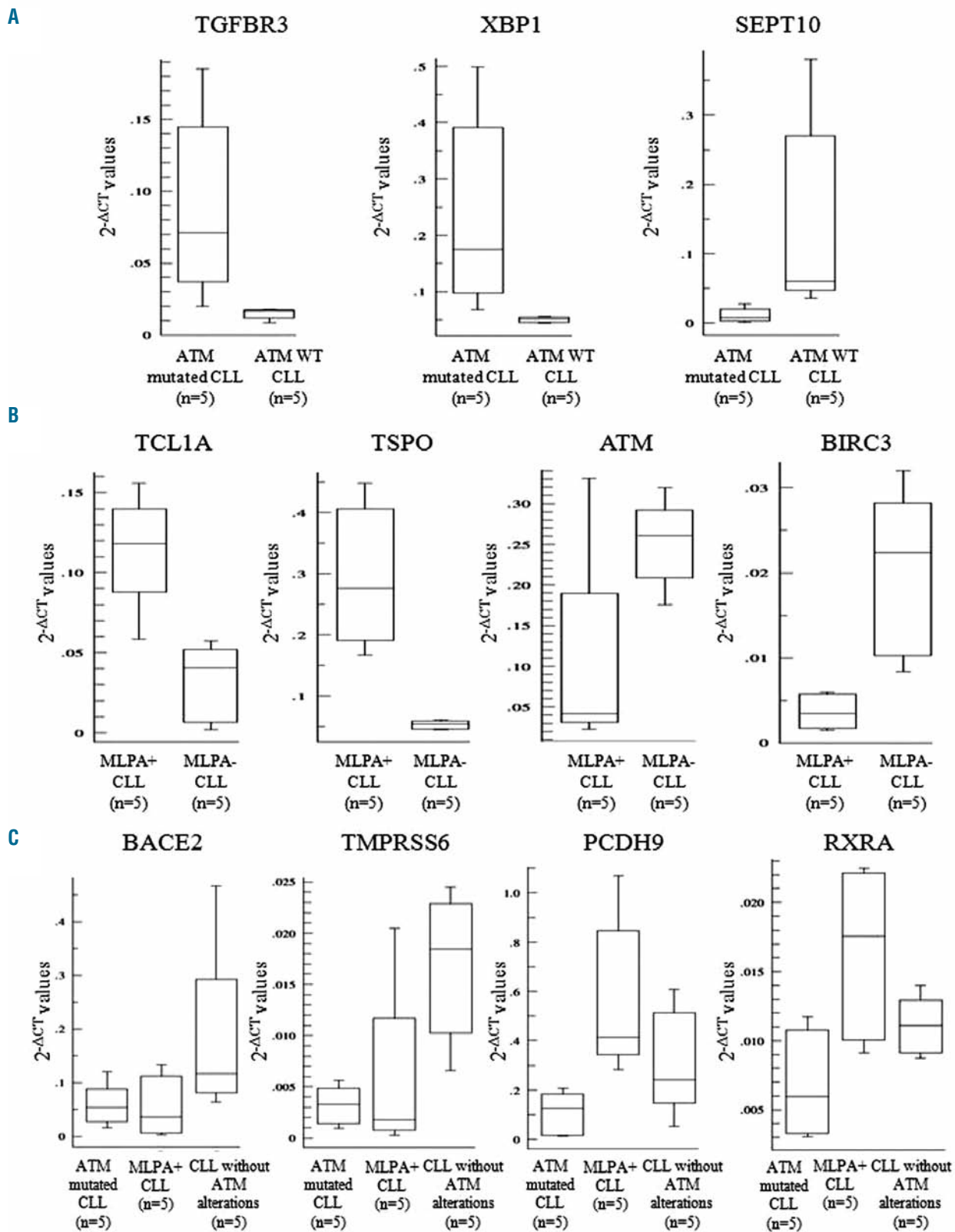
      ↓      ↓
: *** . * : * * * : * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * *
ATM IQSFKAEFRLAGGVNLPKIIDCVGSD-----GKERQQLVKGRRDDLQDAVMQQVFQMCNT
PI3K GALVIEKCKVMASKKKPLWLEFKCADPTALSNETIGIIFKHGDDLQDMLILQILRIMES
. : . : . : . : * : : : : : : : : * * * * * * * * * * * * * * * * * * * * * *
***: ** : * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * *
ATM LLQRNTETRKRKLTICTYKVVPLSQRSGVLEICTGTVPIGEFLVMNEDGAHKRYRPNDFS
PI3K IWET----ESLDLCLLPYGCISFGDKIGMIEIVKDATTIAKIQQSTVGN TGAFKDEV---
: : . * : . * : . . . * : * * . . . . * : : * . . . . :

. * : * * * * . * . * : * * : * * * * * * * * * * * * * * * * * * * * * * *
ATM AFQCQKKMMEVQKKSFEKYEYVFMVDCQNFQPVFRYFCMEKFLDPAIWF EKRLAYTRVA
PI3K -----LSHWLKEKCPLEEFQA AVERFVYS CA
: : : * * : : : . * *

* * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * *
ATM TSSIVGYILGLGDRHVQNILINEQSAELVHIDLGVAFEQGKIL--PTPETVPPFRLTRDIV
PI3K GYCVATFVLGIGDRHNDNIMISE-TGNL FHI DF GHILGNYSFLGINKERVPFVLT P DFL
. : . : * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * *
* * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * *
ATM DGMGITGVEGV-----FRRCEKTMEVVRNSQETLLTIVEVLL
PI3K FVMGTS GKKKTS LHFQK FQDVCVKAYLALRHHINLLIILFSMML
* * : * : * . * * : . * : : * : : * : . . . : *

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Online Supplementary Figure S1. ATM alignment. Sequence alignment of ATM (amino acids 2623-2953, Swiss-Prot entry Q13315) and porcine PI3K γ (amino acids 738-1035, Swiss-Prot entry O02697) employed in the homology modeling of the ATM kinase domain. α -helix and β -sheet secondary structures are indicated respectively by the red and blue colours of the amino acid letters. Aligned amino acids of the two proteins that share the same secondary structure are enclosed in boxes. The amino acids composing the nucleotide binding loop are enclosed in the gray oval. Active site lysine and aspartic acid residues are highlighted by green ovals. Sites of mutations on ATM are indicated by the arrows. Conservation across ATM species is reported above the sequence alignment, while conservation between ATM and porcine PI3K γ is reported just below the same alignment (invariant, conserved and semiconserved residues are indicated, respectively, by an asterisk, a colon and a period). ATM conservation across species was derived from the alignment of ATM sequences from Swiss-Prot database: *Homo sapiens* (Q13315), *Danio rerio* (Q59IS5), *Xenopus laevis* (Q5MPF8), *Mus musculus* (Q62388), *Sus scrofa* (Q6PQD5).



Online Supplementary Figure S2. Validation by Q-PCR method of significantly down- or up-modulated transcripts identified by microarray analysis. (A) Differentially expressed genes in *ATM* mutated patients compared to *ATM* cases without alterations; (B) Differentially expressed genes in MLPA+ patients compared to MLPA- patients; (C) Differentially expressed genes in both *ATM* mutated and MLPA+ patients compared to cases without *ATM* alterations. Gene expression values are expressed as $2^{-\Delta CT}$ values. Box plots define the median values, 25 to 75% of values around the median and the range of values.

Online Supplementary Table S1. Biological characteristics of the 57 CLL patients studied for ATM mutations.

Parameters	N. of patients studied (%)
CD38 expression	57
>7%	17 (30%)
<7%	40 (70%)
ZAP-70 expression	53
>20%	22 (42%)
<20%	31 (58%)
11q22.3 deletion	57
>5%-10%	4 (7%)
>10%	10 (18%)
17p13.1 deletion	57
>5%-20%	8 (14%)
>20%	2 (4%)
13q14 deletion	57
>5%	39 (68%)
14q32 deletion	57
>5%	16 (28%)
Trisomy 12	57
>5%	4 (7%)
IGHV mutation status	57
Unmutated	28 (49%)
Mutated	29 (51%)
TP53	57
Mutated	3 (5%)

Online Supplementary Table S2. Gene symbols and primers evaluated by Q-PCR analysis.

Gene symbol	Forward primer	Reverse primer
<i>GAPDH</i>	CCACCCATGGCAAATTC	GATGGGATTTCCATTGATGACA
<i>ATM</i>	AAATTTTCAACCAGTTTTCCGTTACTT	ACACTGCGCGTATAAGCCAAT
<i>BACE2</i>	CGAGCCCCGTGCAGAAAT	AGTTGTGGCTACATCCTCTGTT
<i>BIRC3</i>	TTTCCGTGGCTCTTATTCAAAC	CTTCTCATCAAGGCAGAAAAATCTT
<i>PCDH9</i>	GCTTGTGCTTGTATTCTTTATGTTAA	CTCCATAGTCTGCGGATCAA
<i>RXRA</i>	AAGGACCGGAACGAGAATGA	ATCCTCTCCACCGGCATGT
<i>SEPT10</i>	ACAGTGGGATTTGGTGACCAA	GGCCTCAAACCTGAGCATCTATGT
<i>TCLIA</i>	GCCTGGGAGAAGTTCGTGTATT	CTGTAACCTATCCTTTATCTCGATGGT
<i>TGFBR3</i>	TGCCAGAGAATGGACACGTTTA	AGCACGTTTGGATGGCAAA
<i>TMPRSS6</i>	GCCACATTCAGTGCAAAGA	CGCTGCCGTTGAGACAATC
<i>TSPO</i>	TGGAAGAGCTGGGAGGCTT	TGTCGGGCACCAAGAAGAT
<i>XBPI</i>	TCTCAGCCCCTCAGAGAATGAT	TCCGGAACGAGGTCATCTTCTA

Online Supplementary Table S3. ATM gene variants and polymorphisms detected in CLL patients.

ID Number	Patients	ATM gene variants					Reference [§]
		Nucleotide Change	Amino acid Change	Type	Exon/Intron	Allelic* Frequency Controls (%)	
5394	C.C.	IVS62+8A>C	—	Intronic	62	14	Castellvi-Bel <i>et al.</i> ¹
3707	B.P.	IVS62+8A>C	—	Intronic	62		
5704	P.A.	IVS14-55T>G	—	Intronic	15	8	Castellvi-Bel <i>et al.</i> ¹
5699	R.D.	IVS14-55T>G	—	Intronic	15		
3442	S.A.	IVS14-55T>G	—	Intronic	15		
5646	A.E.	IVS14-55T>G	—	Intronic	15	8	Sandoval <i>et al.</i> ²
		5557G>A	D1853N	Missense	39	3	
3459	S.A.	5557G>A	D1853N	Missense	39	3	Sandoval <i>et al.</i> ²
3469	R.B.	5558A>T	D1853V	Missense	39	1	Sandoval <i>et al.</i> ² Schaffner <i>et al.</i> ³
3580	C.M.	IVS38-8T>C	—	Intronic	39		—
5281	B.A.	IVS38-8T>C	—	Intronic	39	3	Meier <i>et al.</i> ⁴
		2119T>C	S707P	Missense	15		Koinuma <i>et al.</i> ⁵
3668	S.L.	IVS57+90G>A	—	Intronic	57	1	—
3751	D.C.	5748C>T	D1914D	Silent	40	1	—
3458	S.M.G	5748C>T	D1914D	Silent	40		—
3664	M.R.	IVS55+79T>C	—	Intronic	55	0	#

*180 healthy control subjects; [§]ATM gene variants and polymorphisms reported in the literature; [#]<http://www.vmresearch.org/atm.htm>.

References to Online Supplementary Table S3

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5. Koinuma K, Yamashita Y, Liu W, Hatanaka H, Kurashina K, Wada T, et al. Epigenetic silencing of AXIN2 in colorectal carcinoma with microsatellite instability. *Oncogene.* 2006;25 (1):139-46.

Online Supplementary Table S4. Differentially expressed genes between CLL patients with and without *ATM* gene point mutations. Genes are rank-ordered according to their *P* value.

Probeset ID	Gene Symbol	<i>P</i> value	Chromosomal Location	Gene Function	Expression in <i>ATM</i> -Mutated Cases
224795_x_at	<i>IGK</i>	0.000767	2p12	Immune response	High
214836_x_at	<i>IGKC</i>	0.00095	2p12	Immune response	High
211787_s_at	<i>EIF4A</i>	0.003262	17p13	Translation	High
221651_x_at	<i>IGK</i>	0.007507	2p12	Immune response	High
1569110_x_at	<i>LOC728613</i>	0.007568	5p15.33	Unknown	High
221671_x_at	<i>IGK</i>	0.007701	2p12	Immune response	High
222443_s_at	<i>RBM8A</i>	0.012148	1q12	RNA processing	High
201859_at	<i>SRGN</i>	0.017273	10q22.1	Apoptosis	High
204929_s_at	<i>VAMP5</i>	0.018839	2p11.2	Vesicle-mediated transport	High
200670_at	<i>XBPI</i>	0.02972	22q12	Regulation of transcription, DNA-dependent	High
226625_at	<i>TGFBR3</i>	0.045595	1p33-p32	Signal transduction	High
1563473_at	Unknown	0.047765	Unknown	Unknown	High
210150_s_at	<i>LAMA5</i>	0.000084	20q13.2-q13.3	Angiogenesis	Low
231735_s_at	<i>MALAT1</i>	0.000906	11q13.1	Unknown	Low
239369_at	<i>LCN8</i>	0.002436	9q34.3	Phospholipid metabolic process	Low
232471_at	Unknown	0.004535	Unknown	Unknown	Low
217853_at	<i>TNS3</i>	0.006553	7p12.3	Intracellular signaling cascade	Low
205859_at	<i>LY86</i>	0.008207	6p25.1	Immune response	Low
219737_s_at	<i>PCDH9</i>	0.011019	13q14.3-q21.1	Cell adhesion	Low
1557122_s_at	<i>GABRB2</i>	0.011915	5q34	Ion transport	Low
206206_at	<i>CD180</i>	0.012872	5q12	Immune response	Low
224823_at	<i>MYLK</i>	0.013742	3q21	Protein amino acid phosphorylation	Low
222446_s_at	<i>BACE2</i>	0.014523	21q22.3	Proteolysis	Low
213502_x_at	<i>LOC91316</i>	0.016196	22q11.23	Carbohydrate metabolic process	Low
202449_s_at	<i>RXRA</i>	0.017499	9q34.3	Regulation of transcription, DNA-dependent	Low
217867_x_at	<i>BACE2</i>	0.020668	21q22.3	Proteolysis	Low
235522_at	<i>CLEC2D</i>	0.020993	12p13	Cell surface receptor linked signal transduction	Low
212698_s_at	<i>SEPT10</i>	0.02323	2q13	Cell cycle	Low
222073_at	<i>COL4A3</i>	0.023512	2q36-q37	Cell surface receptor linked signal transduction	Low
217950_at	<i>NOSIP</i>	0.024734	19q13.33	Negative regulation of nitric-oxide synthase activity	Low
222696_at	<i>AXIN2</i>	0.024758	17q23-q24	Signal transduction	Low
209829_at	<i>C6orf32</i>	0.030694	6p22.3-p21.32	Multicellular organismal development	Low
209469_at	<i>GPM6A</i>	0.03877	4q34	Unknown	Low
234367_x_at	<i>TMPRSS6</i>	0.04024	22q12.3	Intracellular signaling cascade	Low
212592_at	<i>IGJ</i>	0.042982	4q21	Immune response	Low

Online Supplementary Table S5. Differentially expressed genes between MLPA+ and MLPA- CLL patients. Genes are rank-ordered according to their P value.

Probeset ID	Gene Symbol	P value	Chromosomal Location	Gene Function	Expression in MLPA+ Cases
209995_s_at	<i>TCLIA</i>	0.000197	14q32.1	Multicellular organismal development	High
39318_at	<i>TCLIA</i>	0.000425	14q32.1	Multicellular organismal development	High
202449_s_at	<i>RXRA</i>	0.001776	9q34.3	Regulation of transcription, DNA-dependent	High
228476_at	<i>KIAA1407</i>	0.00429	3q13.31	Unknown	High
244740_at	<i>MGC9913</i>	0.004904	19q13.43	Unknown	High
210949_s_at	<i>EIF3C</i>	0.005208	16p11.2	Regulation of translational initiation	High
219737_s_at	<i>PCDH9</i>	0.005278	13q14.3-q21.1	Cell adhesion	High
203454_s_at	<i>ATOX1</i>	0.005982	5q32	Cellular copper ion homeostasis	High
200647_x_at	<i>EIF3C</i>	0.006259	16p11.2	Regulation of translational initiation	High
229344_x_at	<i>FAM80B</i>	0.006455	12p13.31	Protein modification process	High
210401_at	<i>P2RX1</i>	0.009813	17p13.3	Signal transduction	High
221253_s_at	<i>TXNDC5</i>	0.009822	6p24.3	Anti-apoptosis	High
215440_s_at	<i>BEX4</i>	0.009915	Xq22.1-q22.3	Unknown	High
215230_x_at	<i>EIF3C</i>	0.010132	16p11.2	Regulation of translational initiation	High
238919_at	Unknown	0.011614	Unknown	Unknown	High
226164_x_at	<i>FAM80B</i>	0.011676	12p13.31	Protein modification process	High
203881_s_at	<i>DMD</i>	0.013194	Xp21.2	Cytoskeletal anchoring	High
1552807_a_at	<i>SIGLEC10</i>	0.015718	19q13.3	Cell adhesion	High
202180_s_at	<i>MVP</i>	0.021829	16p13.1-p11.2	Protein transport	High
218243_at	<i>RUFY1</i>	0.021861	5q35.3	Protein transport	High
235372_at	<i>FCRLA</i>	0.022754	1q23.3	Cell differentiation	High
203028_s_at	<i>CYBA</i>	0.024543	16q24	Superoxide metabolic process	High
213674_x_at	<i>IGHD</i>	0.027414	14q32.33	Immune response	High
201518_at	<i>CBX1</i>	0.028095	17q	Chromatin assembly or disassembly	High
223207_x_at	<i>PHPT1</i>	0.028239	9q34.3	Dephosphorylation	High
219359_at	<i>ATHL1</i>	0.028367	11p15.5	Carbohydrate metabolic process	High
208741_at	<i>SAPI8</i>	0.031401	13q12.11	Regulation of transcription, DNA-dependent	High
213436_at	<i>CNR1</i>	0.035432	6q14-q15	G-protein coupled receptor protein signaling pathway	High
207713_s_at	<i>RBCK1</i>	0.036438	20p13	Protein modification process	High
214366_s_at	<i>ALOX5</i>	0.037884	10q11.2	Leukotriene metabolic process	High
1560225_at	<i>CNR1</i>	0.039142	6q14-q15	G-protein coupled receptor protein signaling pathway	High
204409_s_at	<i>EIF1AY</i>	0.039817	Yq11.222	Translational initiation	High
202098_s_at	<i>PRMT2</i>	0.040657	21q22.3	Signal transduction	High
200701_at	<i>NPC2</i>	0.041376	14q24.3	Phospholipid transport	High
38671_at	<i>PLXND1</i>	0.041874	3q21.3	Signal transduction	High
201400_at	<i>PSMB3</i>	0.042024	17q12	Ubiquitin-dependent protein catabolic process	High
211395_x_at	<i>FCGR2C</i>	0.04339	1q23.3	Immune response	High
202096_s_at	<i>TSPO</i>	0.043686	22q13.31	Steroid metabolic process	High
214933_at	<i>CACNA1A</i>	0.043866	19p13.2-p13.1	Calcium ion transport	High
202709_at	<i>FMOD</i>	0.046059	1q32	Transforming growth factor beta receptor complex assembly	High
200931_s_at	<i>VCL</i>	0.046774	10q22.1-q23	Cell adhesion	High
219922_s_at	<i>LTBP3</i>	0.047574	11q12	Growth factor binding	High
222245_s_at	<i>FER1L1</i>	0.047758	20q11.22	Unknown	High
214916_x_at	Unknown	0.048092	Unknown	Unknown	High
203531_at	<i>CUL5</i>	0.000005	11q22-q23	Negative regulation of cell proliferation	Low
211967_at	<i>TMEM123</i>	0.000052	11q22.1	Receptor activity	Low
227208_at	<i>CCDC84</i>	0.000106	11q23.3	Unknown	Low
221580_s_at	<i>JOSD3</i>	0.000135	11q21	Protein binding	Low
210538_s_at	<i>BIRC3</i>	0.000364	11q22	Anti-apoptosis	Low
201034_at	<i>ADD3</i>	0.000439	10q24.2-q24.3	Structural constituent of cytoskeleton	Low
206126_at	<i>CXCR5</i>	0.000442	11q23.3	G-protein coupled receptor protein signaling pathway; B cell activation	Low
203642_s_at	<i>COBLL1</i>	0.000554	2q24.3	Cell adhesion	Low
217979_at	<i>TSPAN13</i>	0.000573	7p21.1	Signal transduction	Low

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201753_s_at	<i>ADD3</i>	0.000626	10q24.2-q24.3	Structural constituent of cytoskeleton	Low
1558662_s_at	<i>BANK1</i>	0.000702	4q24	B cell activation	Low
212672_at	<i>ATM</i>	0.000942	11q22-q23	DNA repair	Low
215145_s_at	<i>CNTNAP2</i>	0.000976	7q35-q36	Cell adhesion	Low
219300_s_at	<i>CNTNAP2</i>	0.001104	7q35-q36	Cell adhesion	Low
226247_at	<i>PLEKHA1</i>	0.001461	10q26.13	Phospholipid binding	Low
219667_s_at	<i>BANK1</i>	0.001462	4q24	B cell activation	Low
202265_at	<i>BM11</i>	0.001485	10p11.23	Regulation of transcription, DNA-dependent	Low
222808_at	<i>ALG13</i>	0.001855	Xq23	Carbohydrate metabolic process	Low
209750_at	<i>NR1D2</i>	0.001871	3p24.2	Regulation of transcription, DNA-dependent	Low
222446_s_at	<i>BACE2</i>	0.002378	21q22.3	Proteolysis	Low
201752_s_at	<i>ADD3</i>	0.003297	10q24.2-q24.3	Structural constituent of cytoskeleton	Low
225123_at	Unknown	0.003584	Unknown	Unknown	Low
231839_at	<i>2'-PDE</i>	0.003588	3p14.3	Unknown	Low
222915_s_at	<i>BANK1</i>	0.003882	4q24	B cell activation	Low
205882_x_at	<i>ADD3</i>	0.003983	10q24.2-q24.3	Structural constituent of cytoskeleton	Low
222728_s_at	<i>JOSD3</i>	0.004153	11q21	Protein binding	Low
219301_s_at	<i>CNTNAP2</i>	0.004342	7q35-q36	Cell adhesion	Low
203647_s_at	<i>FDXI</i>	0.004429	11q22	Electron transport; steroid metabolic process	Low
243798_at	Unknown	0.004629	Unknown	Unknown	Low
238587_at	<i>UBASH3B</i>	0.004685	11q24.1	Negative regulation of endocytosis	Low
235626_at	<i>CAMK1D</i>	0.004936	10p13	Protein amino acid phosphorylation	Low
213034_at	<i>KIAA0999</i>	0.005176	11q23.3	Protein amino acid phosphorylation	Low
203544_s_at	<i>STAM</i>	0.00541	10p14-p13	Signal transduction	Low
206194_at	<i>HOXC4</i>	0.005749	12q13.3	Regulation of transcription, DNA-dependent	Low
204912_at	<i>IL10RA</i>	0.005817	11q23	Receptor activity	Low
202600_s_at	<i>NR1P1</i>	0.005833	21q11.2	Regulation of transcription, DNA-dependent	Low
224777_s_at	<i>PAFAH1B2</i>	0.005932	11q23	Lipid metabolic process	Low
222792_s_at	<i>CCDC59</i>	0.00599	12q21.31	Regulation of transcription, DNA-dependent	Low
209191_at	<i>TUBB6</i>	0.007009	18p11.21	Microtubule-based process	Low
1568249_at	<i>SNORA71B</i>	0.007033	20q11.23	Unknown	Low
202038_at	<i>UBE4A</i>	0.008139	11q23.3	Ubiquitin-dependent protein catabolic process	Low
212119_at	<i>RHOQ</i>	0.009427	2p21	Small GTPase mediated signal transduction	Low
204621_s_at	<i>NR4A2</i>	0.009608	2q22-q23	Regulation of transcription, DNA-dependent	Low
207826_s_at	<i>ID3</i>	0.009871	1p36.13-p36.12	Negative regulation of transcription	Low
212080_at	<i>MLL</i>	0.010466	11q23	Regulation of transcription, DNA-dependent	Low
209081_s_at	<i>COL18A1</i>	0.010766	21q22.3	Negative regulation of cell proliferation	Low
235739_at	Unknown	0.011077	Unknown	Unknown	Low
225768_at	<i>NR1D2</i>	0.01231	3p24.2	Regulation of transcription, DNA-dependent	Low
210279_at	<i>GPR18</i>	0.012375	13q32	G-protein coupled receptor protein signaling pathway	Low
228528_at	Unknown	0.0124	Unknown	Unknown	Low
226981_at	<i>MLL</i>	0.012427	11q23	Regulation of transcription, DNA-dependent	Low
237040_at	<i>CWF19L2</i>	0.013473	11q22.3	Unknown	Low
229390_at	<i>FAM26F</i>	0.013655	6q22.1	Unknown	Low
230499_at	Unknown	0.014818	Unknown	Unknown	Low
219228_at	<i>ZNF331</i>	0.016173	19q13.41	Regulation of transcription, DNA-dependent	Low
212076_at	<i>MLL</i>	0.016895	11q23	Regulation of transcription, DNA-dependent	Low
224642_at	<i>FYTTD1</i>	0.01713	3q29	Unknown	Low
204622_x_at	<i>NR4A2</i>	0.017188	2q22-q23	Regulation of transcription, DNA-dependent	Low
210258_at	<i>RGS13</i>	0.017358	1q31.2	G-protein coupled receptor protein signaling pathway	Low
234367_x_at	<i>TMPRSS6</i>	0.017908	22q12.3	Intracellular signaling cascade	Low
226763_at	<i>SESTD1</i>	0.018889	2q31.2	Unknown	Low
218750_at	<i>JOSD3</i>	0.019124	11q21	Protein binding	Low
235170_at	<i>ZNF92</i>	0.020987	7q11.21	Regulation of transcription, DNA-dependent	Low

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242920_at	Unknown	0.022263	Unknown	Unknown	Low
216834_at	<i>RGS1</i>	0.023097	1q31	Immune response; G-protein signaling, adenylate cyclase inhibiting pathway	Low
205419_at	<i>EBI2</i>	0.024464	13q32.3	Immune response; G-protein coupled receptor protein signaling pathway	Low
231124_x_at	<i>LY9</i>	0.025138	1q21.3-q22	Immunoglobulin mediated immune response	Low
230128_at	<i>IGL</i>	0.02922	22q11.1-q11.2	tRNA aminoacylation for protein translation	Low
216248_s_at	<i>NR4A2</i>	0.029801	2q22-q23	Regulation of transcription, DNA-dependent	Low
217867_x_at	<i>BACE2</i>	0.031937	21q22.3	Proteolysis	Low
225954_s_at	<i>MIDN</i>	0.038338	19p13.3	Protein modification process	Low
227189_at	<i>CPNE5</i>	0.042492	6p21.1	Signal transduction	Low
233952_s_at	<i>ZNF295</i>	0.043895	21q22.3	Regulation of transcription, DNA-dependent	Low
201525_at	<i>APOD</i>	0.048048	3q26.2-qter	Lipid metabolic process	Low