

Inflammatory cytokines and signaling pathways are associated with survival of primary chronic lymphocytic leukemia cells *in vitro*: a dominant role of CCL2

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Supplementary Discussion

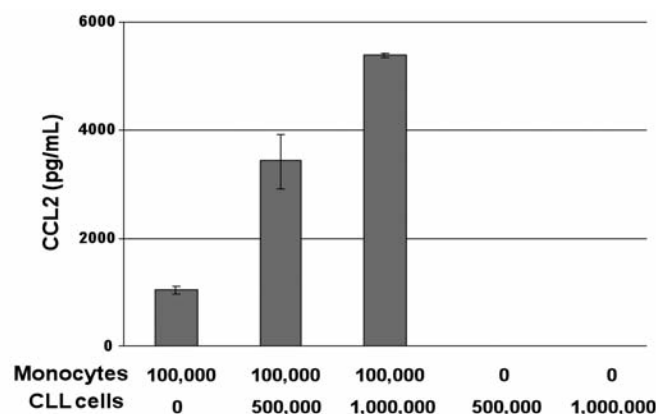
Cytokine antibody array results

To study the composition of soluble factors in CLL/HS-5 co-cultures, we performed cytokine antibody arrays using supernatants of such co-cultures and pure HS-5 control cultures. The most abundant cytokines in the co-cultures included interleukin-6 and interleukin-8, which are both known to be survival-inducing factors for CLL cells.^{1,2} We also detected high levels of CCL4 and weaker expression of CCL3 in a subset of CLL patients, which is in accordance with published data describing expression of these two T-cell chemokines in CLL/nurse-like cell co-cultures.³ In addition, we identified the expression of chemokines which have not been associated with CLL so far, including CCL2, CCL7, CXCL1, and CXCL5, all of which are known to have chemotactic activity for leukocytes.⁴ Further novel genes in relation to CLL were the two matrix metallopro-

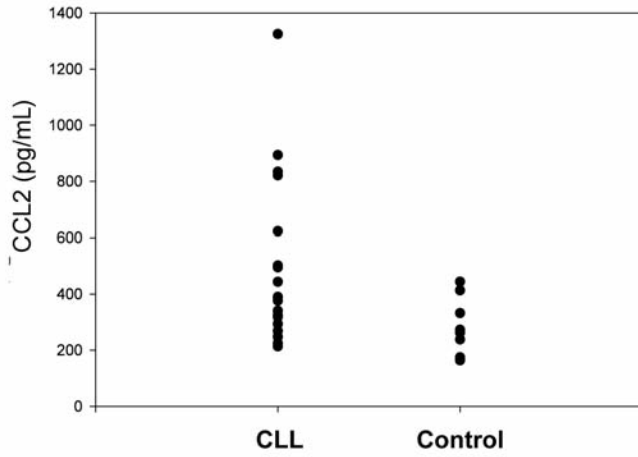
teinases MMP1 and MMP3 and their inhibitors TIMP1 and TIMP2, which are known to be transcriptionally regulated by a variety of cytokines. Leukemia inhibitory factor (LIF), which was among the top 15 candidates in the cytokine arrays, was recently shown to be a regulator of MMP1, MMP3, and TIMP1 expression.⁵ Besides their ability to degrade extracellular matrix, MMP can also process cell surface receptors and bioactive factors, such as cytokines and chemokines. By this means, MMP and their regulators are important mediators of cell migration and inflammatory processes.⁶ Several MMP have been associated with the development and progression of a variety of tumors, including CLL and there are a number of MMP inhibitors that are currently being tested in all three phases of clinical trials against a variety of human cancers.⁷ The potential relevance of the identified MMP and their regulators in CLL will have to be analyzed in future functional studies using our *in vitro* model.

References

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Online Supplementary Figure S1. Increased CCL2 secretion by monocytes in the presence of CLL cells is not due to smaller numbers of monocytes present in the CLL cell preparations. 1×10^5 CD14-sorted monocytes were cultured in the absence or presence of 5×10^5 or 1×10^6 CD19-sorted CLL cells in 250 μ L complete medium per well in 24-well plates for 2 days. In addition, 5×10^5 or 1×10^6 CD19-sorted CLL cells were cultured without monocytes. Cell culture supernatants were analyzed by ELISA for the presence of CCL2. Mean values and standard deviations of duplicates of one representative example out of two independently performed experiments are shown.



Online Supplementary Figure S2. Blood serum levels of CCL2 were quantified in 21 CLL patients and ten healthy controls by ELISA. Single values of all samples analyzed are depicted in the dot blot.

Online Supplementary Table S1. Patients' data, including clinical stage at time of investigation, fluorescence *in situ* hybridization (FISH) results, mutational status, *VH* gene usage, ZAP-70 results, and assays performed.

Patient ID Viable cells	Clinical stage	FISH	Mutational status	<i>V_H</i> gene usage	ZAP-70	Assays performed
CLL 1	C	normal	UM	V3-23	pos	DNA Microarray Cocu
CLL 3	C	11q-, 13q-	UM	V1-69	neg	DNA Microarray Cocu
CLL 6	A	13q-	M, out of frame	V3-43	neg	DNA Microarray Cocu
CLL 7	C	normal	M	V4-34	neg	DNA Microarray Cocu
CLL 8	B	13q-	M	V1-81	nd	CCL2 ELISA
CLL 15	A	13q-	M	V3-23	neg	qRT-PCR
CLL 16	A	13q-	UM	V1-69	neg	qRT-PCR
CLL 17	A	normal	M	V3-66	neg	qRT-PCR
CLL 18	A	13q-	M	V4-39	neg	qRT-PCR
CLL 19	A	13q-	M	V5-51	neg	qRT-PCR
CLL 23	A	normal	UM	V1-01	nd	CCL2 ELISA
CLL 28	C	13q-	M	V4-34	neg	qRT-PCR (CD19+)
CLL 29	A	13q-	M	V1-69	neg	qRT-PCR (CD19+)
CLL 30	B	17p-	UM	V3-30	neg	qRT-PCR (CD19+)
CLL 43	A	13q-	M	DP58/hv3d1EG	neg	qRT-PCR
CLL 44	B	13q-	nd	nd	pos	DNA Microarray CM; qRT-PCR
CLL 45	B	13q-	nd	nd	neg	DNA Microarray CM; qRT-PCR
CLL 46	B	11q-, 13q-, 14q-	UM	V1-69	neg	DNA Microarray CM
CLL 48	A	13q-	M	V1-18	neg	DNA Microarray CM
CLL 49	B	normal	UM	V1-02	neg	DNA Microarray CM
CLL 58	A	normal	UM	V3-33	pos	DNA Microarray CD; Cytokine Array; CCL2 ELISA
CLL 59	A	normal	M	V3-21	pos	DNA Microarray CD
CLL 60	A	13q-	M	V3-07	neg	DNA Microarray CD; Cytokine Array
CLL 61	C	11q-, 13q-	UM	V3-11	neg	DNA Microarray CD; Cytokine Array
CLL 64	B	13q-	M	V1-69	neg	CCL2 ELISA
CLL 67	B	13q-	M	V1-81	nd	CCL2 qRT-PCR + ELISA
CLL 68	A	normal	UM	V3-33	pos	CCL2 qRT-PCR
CLL 70	B	13q-	M	V3-53	neg	Survival Cocu, CM, CD
CLL 72	C	13q-	M	V4-34	neg	Survival Cocu, CM, CD
CLL 74	A	13q-	UM	V1-69	neg	Survival Cocu, CM, CD
CLL 82	A	normal	UM	V3-33	pos	CCL2 qRT-PCR

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Patient ID Serum	Clinical stage	FISH	Mutational status	V _H gene usage	ZAP-70	Assays performed
CLL S1	A	normal	M	V4-61	nd	CCL2 ELISA
CLL S2	A	t(6;12), 13q-	M	V4-59	nd	CCL2 ELISA
CLL S3	A	13q-	M	V4-59	neg	CCL2 ELISA
CLL S4	A	13q-	M	V3-23	neg	CCL2 ELISA
CLL S5	A	normal	M	V3-53	neg	CCL2 ELISA
CLL S6	A	normal	UM	V1-69	pos	CCL2 ELISA
CLL S7	A	normal	UM	V4-59	pos	CCL2 ELISA
CLL S8	nd	normal	UM	V3-48	nd	CCL2 ELISA
CLL S9	C	normal	UM	V3-23	neg	CCL2 ELISA
CLL S10	B	13q-, 11q-	UM	V1-69	nd	CCL2 ELISA
CLL S11	A	13q-	M	V3-72	nd	CCL2 ELISA
CLL S12	nd	13q-	UM	V3-30	pos	CCL2 ELISA
CLL S13	A	13q-	M	V3-21	neg	CCL2 ELISA
CLL S14	A	11q-, 13q-	UM	nd	nd	CCL2 ELISA
CLL S15	A	normal	M	V4-30.4	neg	CCL2 ELISA
CLL S16	A	normal	M	V4-34	nd	CCL2 ELISA
CLL S17	A	normal	M	V1-08	nd	CCL2 ELISA
CLL S18	nd	normal	M	nd	nd	CCL2 ELISA
CLL S19	A	13q-	M	V3-34	nd	CCL2 ELISA
CLL S20	A	11q-, 13q-	M	V3-21	neg	CCL2 ELISA
CLL S21	A	normal	M	V3-66	neg	CCL2 ELISA

UM: unmutated; M: mutated; nd: not determined.

Online Supplementary Table S2. Sequences of primers used for qRT-PCR.

Gene name	Forward primer	Reverse primer
<i>BRWD1</i>	ctcatcgagtcggagctg	tccgcaacaactggctactg
<i>CCL2</i>	agtctctgccgcccttct	gtgactggggcattgattg
<i>CCL22</i>	ggcctggcgtggtgtgctaacttc	cagggccttcacagtttgat
<i>CD180</i>	cagggccttcacagtttgat	acatctgatcccaggagggtg
<i>CDK2AP2</i>	agatccggcctacctatgct	gataggtccagacgctgagg
<i>DCTN2</i>	actagcgacctacctgagga	tcataggcagcattaggattgac
<i>EGLN2</i>	tgggcagctatgtcatcaac	aatgagcaaccggtcaaag
<i>HPRT</i>	aattatggacaggactgaactctgct	tccagcaggtcagaaagaattatagc
<i>LIMS3</i>	atgagcagtggttcgtgtgc	cccttgactcagcaccagt
<i>MYCN</i>	gactgtagccatccgaggac	gttttaataccgggggtgct
<i>NFKBIA</i>	aacctgcagcagactccact	gacacgtgtggccattgtag
<i>PARP9</i>	tagctctccagcttccctcg	tggaaaagtcctcctcag
<i>PBGD</i>	tccagagaagagtggtg	aagccgggtgttgaggtt
<i>PGK1</i>	aagtgaagctcgaaagcttctat	tgggaaaagatgcttctggg
<i>PP1A</i>	ccgagactggaagacaggag	cccacctttccttccatt
<i>RB1CC1</i>	cagtgcctgacgtaactgtatc	gtctgccacagtttgac
<i>RHOB</i>	cgacgtcattctcatgtct	cgaggtagtcgtaggcttg
<i>SDHA</i>	ggacctggtgtctttggtc	ccagcgtttggtttaattgg
<i>SNIP1</i>	aaggcctcaggagaagaag	ccggtcactgtcctagctc
<i>SPDYA</i>	gaagagtgccagacaatg	caaagctactgcgtcaaatg

Online Supplementary Table S3. Top differentially expressed genes which are unique to CLL cells in HS-5 co-culture. The columns of the table show the gene symbols, the chromosomal mapping of the genes (Mapping), log₂ fold expression changes (log FC), average log₂ expression (Ave Expr), moderated t-statistics (t), raw and adjusted P values (adj. P value), and log odds values (B).

Gene symbols	Mapping	logFC	Ave Expr	t	P value	adj. P value	B
<i>EIF5A</i>	17p13.1	2.35	12.06	31.82	3.14E-22	3.62E-20	40.92
<i>MX1</i>	21q22.3	1.52	11.55	18.53	2.00E-16	3.21E-15	27.30
<i>STARD10</i>	11q13.4	1.49	10.66	21.27	7.03E-18	1.69E-16	30.73
<i>BCL2L12</i>	19q13.33	1.48	10.72	12.38	2.36E-12	1.46E-11	17.67
<i>C20orf43</i>	20q13.31	1.45	13.27	15.58	1.22E-14	1.25E-13	23.08
<i>NME2P1;NME1-NME2</i>	17q21.33	1.40	13.15	14.79	4.12E-14	3.65E-13	21.83
<i>MAP4K4</i>	2q11.2	1.37	11.77	20.61	1.52E-17	3.31E-16	29.94
<i>TP53</i>	17p13.1	1.29	9.53	18.65	1.72E-16	2.82E-15	27.46
<i>NUTF2</i>	16q22.1	1.28	11.24	14.98	3.07E-14	2.81E-13	22.13
<i>IGFBP6</i>	12q13.13	1.26	10.60	11.39	1.45E-11	7.71E-11	15.81
<i>CRYZ</i>	1p31.1	1.25	10.71	21.60	4.82E-18	1.21E-16	31.12
<i>PLK1</i>	16p12.1	1.24	13.24	14.10	1.24E-13	9.88E-13	20.70
<i>PSMD9</i>	12q24.31	1.22	10.97	13.54	5.89E-13	4.03E-12	19.15
<i>NARS2</i>	11q14.1	1.22	10.70	14.32	8.72E-14	7.21E-13	21.06
<i>COL7A1</i>	3p21.31	1.21	10.89	13.34	8.16E-13	5.43E-12	18.81
<i>GOLIM4</i>	3q26.2	1.20	9.23	17.54	1.73E-15	2.17E-14	25.15
<i>GLRA2</i>	Xp22.2	1.19	10.78	16.18	2.42E-14	2.27E-13	22.49
<i>KIF11</i>	10q23.33	1.19	11.70	11.05	4.53E-11	2.20E-10	14.68
<i>ATL3</i>	11q13.1	1.19	10.60	13.58	2.91E-13	2.13E-12	19.82
<i>FIBP</i>	11q13.1	1.19	13.36	13.25	9.47E-13	6.22E-12	18.66
<i>RB1CC1</i>	8q11.23	-1.19	11.20	-15.21	4.43E-14	3.90E-13	21.81
<i>NFKBIA</i>	14q13.2	-1.19	13.10	-15.41	1.58E-14	1.56E-13	22.82
<i>UBXN7</i>	3q29	-1.20	9.76	-22.88	1.17E-18	3.61E-17	32.56
<i>AC069234.35-201</i>	12q24.31	-1.21	9.96	-17.20	1.19E-15	1.55E-14	25.47
<i>DENND4B</i>	1q21.3	-1.21	10.73	-23.00	9.18E-18	2.11E-16	30.58
<i>NARG1L</i>	13q14.11	-1.21	11.19	-18.85	1.32E-16	2.24E-15	27.73
<i>KIAA1310</i>	2q11.2	-1.22	10.56	-11.43	1.37E-11	7.29E-11	15.87
<i>PHF17</i>	4q28.2	-1.22	12.00	-15.59	1.21E-14	1.25E-13	23.09
<i>USP7</i>	16p13.2	-1.22	10.19	-13.39	4.05E-13	2.88E-12	19.48
<i>POLI</i>	18q21.2	-1.27	10.15	-15.11	2.53E-14	2.36E-13	22.34
<i>PNPLA2</i>	11p15.5	-1.27	12.12	-11.45	1.30E-11	6.95E-11	15.92
<i>NA</i>	6q14.1	-1.28	10.43	-18.18	3.16E-16	4.83E-15	26.83
<i>SUPT7L</i>	2p23.2	-1.28	9.20	-23.98	3.55E-18	9.38E-17	31.56
<i>PABPC1L</i>	20q13.12	-1.29	10.20	-15.33	1.78E-14	1.73E-13	22.69
<i>KLHL24</i>	3q27.1	-1.29	12.32	-15.18	4.65E-14	4.07E-13	21.76
<i>MED13L</i>	12q24.21	-1.33	10.18	-12.28	2.79E-12	1.70E-11	17.50
<i>ARID5B</i>	10q21.2	-1.35	13.24	-20.17	2.57E-17	5.31E-16	29.41
<i>ZMYM2</i>	13q12.11	-1.36	10.01	-13.74	2.25E-13	1.69E-12	20.09
<i>PP1A</i>	6q25.1	-1.36	10.87	-14.75	4.38E-14	3.86E-13	21.77
<i>ZNF44</i>	19p13.2	-1.70	9.45	-20.28	2.26E-17	4.72E-16	29.54

Online Supplementary Table S4. Top differentially expressed genes which are unique to CLL cells cultured in HS-5 conditioned medium. Values are as described in *Online Supplementary Table S3*.

Gene symbols	Mapping	logFC	Ave Expr	t	P value	adj. P value	B
<i>FCRLA</i>	1q23.3	2.19	12.33	25.33	9.43E-20	6.17E-18	35.17
<i>NAPSB</i>	19q13.33	1.93	11.19	16.46	3.35E-15	4.41E-14	24.47
<i>NA</i>	9q31.3	1.89	9.32	23.02	2.99E-18	1.05E-16	31.72
<i>FPR2</i>	19q13.33	1.76	9.54	25.58	2.42E-19	1.30E-17	34.27
<i>NUMA1</i>	11q13.4	1.73	11.08	21.50	1.50E-17	4.07E-16	30.07
<i>FCRL3</i>	1q23.1	1.67	12.18	25.34	1.00E-18	4.29E-17	32.89
<i>GYS1</i>	19q13.33	1.67	10.66	25.88	1.82E-19	1.06E-17	34.56
<i>DENND2D</i>	1p13.3	1.64	9.03	31.64	1.45E-21	1.89E-19	39.45
<i>ANXA6</i>	5q33.1	1.64	12.47	45.63	1.92E-25	1.57E-22	48.29
<i>CRIP3</i>	6p21.1	1.60	9.73	23.16	2.43E-17	6.13E-16	29.70
<i>CASC1</i>	12p12.1	1.59	8.62	23.75	1.43E-18	5.76E-17	32.51
<i>FCRL3</i>	1q23.1	1.59	10.63	22.03	8.49E-18	2.53E-16	30.65
<i>TMEM63A</i>	1q42.12	1.57	10.05	30.21	1.16E-21	1.63E-19	39.62
<i>PFTK1</i>	7q21.13	1.55	11.08	31.84	3.09E-22	5.96E-20	40.96
<i>SELL</i>	1q24.2	1.55	11.87	15.49	2.95E-14	2.99E-13	22.30
<i>NCR3</i>	6p21.33	1.54	10.88	24.51	6.75E-19	3.11E-17	33.23
<i>NCF4</i>	22q12.3	1.54	10.03	26.16	1.41E-19	8.75E-18	34.82
<i>TARS2</i>	1q21.2	1.54	9.99	25.21	3.42E-19	1.74E-17	33.92
<i>LASP1</i>	17q12	1.52	11.45	20.75	1.28E-17	3.56E-16	30.17
<i>CAPG</i>	2p11.2	1.52	13.07	25.01	4.15E-19	2.04E-17	33.73
<i>KIAA2018</i>	3q13.2	-1.17	8.04	-22.87	3.20E-16	5.59E-15	27.25
<i>USP36</i>	17q25.3	-1.17	10.32	-16.84	4.42E-15	5.64E-14	24.24
<i>STX16;NPEPL1</i>	20q13.32	-1.20	11.58	-19.44	6.31E-17	1.37E-15	28.54
<i>DCPIA</i>	3p21.1	-1.23	11.43	-23.66	5.13E-19	2.44E-17	33.45
<i>PDE3A;AC112777.7</i>	12p12.2	-1.23	14.88	-14.13	1.19E-13	1.05E-12	20.81
<i>SAT1</i>	Xp22.11	-1.23	12.51	-15.90	7.64E-15	9.06E-14	23.63
<i>CD44</i>	11p13	-1.23	12.50	-10.06	2.04E-10	9.39E-10	13.16
<i>DENND1A</i>	9q33.2	-1.23	10.47	-21.88	3.51E-18	1.21E-16	31.49
<i>SAT1</i>	Xp22.11	-1.24	13.12	-15.62	1.16E-14	1.31E-13	23.20
<i>TAGLN2</i>	1q23.2	-1.25	13.08	-10.32	1.20E-10	5.76E-10	13.71
<i>SNCA</i>	4q22.1	-1.27	11.50	-16.00	6.58E-15	7.93E-14	23.78
<i>NA</i>	7q32.3	-1.28	9.32	-21.37	1.74E-17	4.57E-16	29.92
<i>TNFSF9</i>	19p13.3	-1.34	12.03	-25.49	8.10E-20	5.57E-18	35.33
<i>DIRC2</i>	3q21.1	-1.38	10.60	-17.87	4.76E-16	7.88E-15	26.47
<i>DNAJB1</i>	19p13.12	-1.41	12.78	-11.12	4.00E-11	2.09E-10	14.96
<i>GDAP1</i>	8q21.11	-1.44	9.91	-31.59	3.79E-22	6.67E-20	40.75
<i>RHBDD2</i>	7q11.23	-1.63	11.69	-28.99	3.26E-21	3.85E-19	38.58
<i>LMNA</i>	1q22	-1.68	12.25	-22.21	2.42E-18	8.81E-17	31.87
<i>ACSM3</i>	16p12.2	-1.83	9.68	-48.86	5.90E-27	8.49E-24	51.68
<i>TNF</i>	6p21.33	-2.42	8.61	-64.11	2.54E-26	2.68E-23	49.99

Online Supplementary Table S5. Top differentially expressed genes which are unique to CLL cells in high cell density culture. Values are as described in *Online Supplementary Table S3*.

Gene symbols	Mapping	logFC	Ave Expr	t	P value	adj. P value	B
<i>PLA2G7</i>	6p12.3	2.29	9.78	45.29	2.30E-25	5.77E-23	48.19
<i>GIMAP5</i>	7q36.1	2.28	9.35	36.28	2.42E-22	1.66E-20	41.28
<i>ZNF33B;ZNF33A</i>	10q11.21	2.23	12.11	30.35	1.03E-21	5.49E-20	39.66
<i>CABLES1</i>	18q11.2	2.21	12.03	18.38	2.43E-16	2.85E-15	26.99
<i>THAP2</i>	12q21.1	2.20	7.93	15.67	5.49E-13	3.28E-12	19.45
<i>ID3</i>	1p36.12	2.15	13.24	29.65	1.86E-21	9.20E-20	39.06
<i>BID</i>	22q11.21	2.11	10.99	37.15	6.29E-24	7.95E-22	44.83
<i>NA</i>	5p13.2	2.11	11.04	29.51	2.10E-21	9.99E-20	38.94
<i>TMEM140</i>	7q33	2.09	9.87	51.87	1.28E-27	8.80E-25	53.31
<i>HVCN1</i>	12q24.11	2.05	12.12	18.88	1.28E-16	1.61E-15	27.65
<i>BCL11A</i>	2p16.1	2.04	10.70	31.99	2.75E-22	1.87E-20	41.00
<i>HMOX1</i>	22q12.3	2.03	10.74	32.42	1.96E-22	1.39E-20	41.35
<i>ZNF613</i>	19q13.33	2.02	8.59	31.81	3.18E-22	2.09E-20	40.86
<i>ATXN1</i>	6p22.3	2.02	11.39	35.78	1.63E-23	1.78E-21	43.87
<i>NLRCA</i>	2p22.3	2.01	9.50	36.73	8.41E-24	1.02E-21	44.54
<i>ZNF322B;ZNF322A</i>	9q22.33	1.95	10.15	31.27	4.89E-22	2.94E-20	40.42
<i>GIMAP7</i>	7q36.1	1.94	9.53	26.88	2.17E-20	7.64E-19	36.55
<i>GPR155</i>	2q31.1	1.93	9.75	23.85	1.29E-18	2.65E-17	32.44
<i>MITD1</i>	2q11.2	1.91	11.66	20.01	3.12E-17	4.58E-16	29.10
<i>NA</i>	3q21.2	1.91	9.13	41.40	1.10E-23	1.30E-21	44.36
<i>CFD</i>	19p13.3	-1.79	8.28	-28.50	2.50E-19	6.32E-18	34.26
<i>RP11-110J1.5;ETV3</i>	1q23.1	-1.79	11.32	-23.83	1.31E-18	2.69E-17	32.42
<i>FAM54A</i>	6q23.3	-1.84	10.47	-33.88	2.75E-22	1.87E-20	41.06
<i>F13A1</i>	6p25.1	-1.84	8.31	-19.89	6.70E-16	7.09E-15	26.34
<i>TWF1</i>	12q12	-1.84	11.74	-27.60	1.12E-20	4.28E-19	37.23
<i>IL6ST</i>	5q11.2	-1.86	11.21	-38.48	2.58E-24	3.75E-22	45.73
<i>ADNP2</i>	18q23	-1.87	10.63	-28.27	6.16E-21	2.57E-19	37.84
<i>OSGIN2</i>	8q21.3	-1.88	9.67	-33.73	7.23E-23	5.89E-21	42.36
<i>CSPG2</i>	5q14.3	-1.91	10.46	-35.04	5.46E-22	3.22E-20	40.46
<i>DDX21</i>	10q21.3	-1.94	11.63	-34.48	4.16E-23	3.89E-21	42.92
<i>KIAA0317</i>	14q24.3	-1.95	10.08	-34.25	4.94E-23	4.46E-21	42.75
<i>SFPQ</i>	1p34.3	-1.95	11.96	-46.95	1.64E-26	7.39E-24	50.79
<i>NA</i>	7p13	-1.95	10.24	-36.37	4.91E-23	4.46E-21	42.81
<i>AMD1</i>	6q21	-1.95	12.06	-34.23	4.98E-23	4.48E-21	42.74
<i>F13A1</i>	6p25.1	-2.01	8.80	-23.72	4.54E-18	8.13E-17	31.29
<i>PBEF1</i>	7q22.2	-2.01	10.80	-37.70	4.32E-24	5.65E-22	45.21
<i>SLC25A33</i>	1p36.22	-2.02	11.93	-40.40	7.49E-25	1.48E-22	46.97
<i>PBEF1</i>	7q22.2	-2.04	13.23	-12.05	4.27E-12	2.22E-11	16.93
<i>SNX3</i>	6q21	-2.09	8.27	-41.11	4.81E-25	1.07E-22	47.42
<i>GEM</i>	8q22.1	-2.21	9.46	-32.05	1.06E-21	5.55E-20	39.69

Online Supplementary Table S6. Percentages and numbers of genes common to all three culture conditions, when analyzing the top 40 fold-change candidates of up- and down-regulated genes in each group.

	Common to all three culture conditions	
	Up-regulated	Down-regulated
CLL/HS-5 co-culture (after depletion of HS-5-specific genes)	63% (25 of 40)	80% (32 of 40)
HS-5 conditioned medium	53% (21 of 40)	85% (34 of 40)
High cell density culture	55% (22 of 40)	93% (37 of 40)

Online Supplementary Table S7. Top differentially expressed genes which are common to the three survival-inducing culture conditions tested (Cocu = HS-5 co-culture; CM = HS-5 conditioned medium; CD = high cell density culture). The columns of the table represent the gene symbols, the chromosomal mapping of the genes, log₂ fold expression changes, average log₂ expression, moderated t-statistics (t), raw and adjusted P-values, and log odds values (B).

GENE SYMBOLS	MAPPING	AveExpr	Cocu logFC	t	P Value	adj.P.Val	B	CM logFC	t	P Value	adj.P.Val	B	CD logFC	t	P Value	adj.P.Val	B
CCL2	17q12	12.05	6.19	26.04	5.33E-19	1.87E-17	33.49	4.46	25.70	7.27E-19	3.30E-17	33.21	4.13	17.35	5.20E-15	4.54E-14	24.12
CTS1	9q21.33	12.35	4.97	32.18	2.37E-22	2.93E-20	41.21	1.22	10.85	4.11E-11	2.14E-10	14.81	1.50	11.90	5.59E-12	2.84E-11	16.66
TPM2	9p13.3	11.41	4.32	39.52	1.31E-24	5.06E-22	46.41	1.43	17.87	4.75E-16	7.88E-15	26.48	1.51	16.93	1.72E-15	1.66E-14	24.98
RAB13;RAB5B	1q21.3	12.22	4.31	68.54	1.02E-30	1.61E-26	59.97	1.24	27.06	1.84E-20	1.66E-18	36.83	1.59	31.02	6.00E-22	3.45E-20	40.21
AL02398.1;TRAF3IP3	1q32.2	10.55	4.29	35.36	2.20E-23	4.46E-21	43.60	1.40	15.84	6.36E-15	9.83E-14	23.53	2.65	26.76	2.43E-20	8.38E-19	36.44
CNNA2	4q27	10.34	4.16	57.06	1.94E-23	4.26E-21	43.76	1.05	17.99	9.89E-14	8.91E-13	21.45	1.03	12.27	1.04E-10	4.34E-10	14.28
S100A11;DPY19L2;AC005400.1;AC007683.5	1q21.3	10.44	3.47	24.93	4.48E-19	1.61E-17	33.61	1.00	9.01	2.70E-09	9.97E-09	10.64	1.37	12.03	7.64E-12	3.80E-11	16.38
CXCL5	16p13.3	13.42	3.41	17.09	1.38E-15	1.77E-14	25.32	2.92	20.00	9.15E-17	7.64E-16	29.25	2.12	13.00	7.84E-13	4.57E-12	18.68
S100A11	1q21.3	12.47	3.41	25.99	5.01E-18	2.40E-18	35.78	1.23	12.86	3.99E-13	7.24E-12	18.62	1.91	17.88	4.73E-16	5.21E-15	26.31
NT5DC2	3p21.1	10.71	3.38	36.63	4.13E-23	7.42E-21	43.01	1.30	17.55	1.72E-15	2.44E-14	25.28	1.14	15.11	5.12E-14	3.68E-13	21.54
RAB23	6p12.1	10.52	3.20	31.81	1.27E-21	1.15E-19	39.56	1.25	15.60	2.52E-14	2.62E-13	22.53	1.03	12.59	2.88E-12	1.53E-11	17.39
UPP1	7p12.3	10.75	3.12	33.39	9.33E-23	1.49E-20	42.14	1.18	17.36	9.50E-16	1.45E-14	25.76	1.04	13.59	2.89E-13	1.81E-12	19.71
SLC16A3	17q25.3	11.37	3.11	27.75	3.44E-20	1.73E-18	36.22	1.00	12.29	4.81E-12	3.02E-11	17.06	1.50	14.64	1.04E-13	7.09E-13	20.91
DSCR1	21q22.12	12.12	2.97	29.11	2.95E-21	2.28E-19	38.66	1.97	26.41	3.34E-20	2.71E-18	36.23	2.16	25.95	5.17E-20	1.61E-18	35.66
C15orf48;SPATA5L1	15q21.1	10.31	2.91	22.19	2.49E-18	6.90E-17	31.80	1.46	15.20	2.19E-14	2.32E-13	22.55	1.84	17.17	1.24E-15	1.24E-14	25.31
LTBP2	14q24.3	10.89	2.91	32.42	1.96E-22	2.61E-20	41.40	1.16	17.74	5.65E-16	9.18E-15	26.30	1.46	19.88	3.67E-17	5.25E-16	28.93
TUBG1	17q21.31	11.88	2.81	24.92	1.42E-19	5.94E-18	34.72	1.19	14.42	7.37E-14	6.81E-13	21.30	1.87	20.33	2.11E-17	3.23E-16	29.50
CCT5	5p15.2	12.99	2.80	24.46	2.26E-19	8.89E-18	34.24	1.18	14.43	7.31E-14	6.76E-13	21.31	1.47	15.72	9.99E-15	8.21E-14	23.17
SLC43A3	11q12.1	11.77	2.78	34.94	2.97E-23	5.79E-21	43.30	1.20	20.22	2.42E-17	6.12E-16	29.52	1.91	29.29	2.53E-21	1.18E-19	38.75
SAC3D1	11q13.1	10.30	2.75	26.91	2.52E-19	9.83E-18	34.26	1.64	21.95	2.67E-17	6.63E-16	29.54	1.85	18.13	1.96E-15	1.88E-14	25.12
AC114982.2;IPO11	5q12.1	8.71	-2.77	-29.48	7.99E-21	5.16E-19	37.70	-2.00	-29.09	1.10E-20	1.11E-18	37.40	-2.31	-30.10	4.83E-21	2.08E-19	38.15
C13orf15	13q14.11	13.00	-2.87	-20.82	1.19E-17	2.65E-16	30.20	-1.89	-18.82	1.38E-16	2.69E-15	27.74	-1.48	-13.16	5.95E-13	3.53E-12	18.96
ZNF394;ZNF789	7q22.1	10.70	-2.91	-43.42	1.19E-25	7.80E-23	48.79	-1.82	-37.29	5.72E-24	2.38E-21	44.94	-2.90	-53.07	7.15E-28	5.14E-25	53.86
NR4A2	2q24.1	10.68	-2.96	-18.92	3.00E-16	4.63E-15	26.95	-2.66	-23.24	2.39E-18	8.70E-17	31.94	-4.43	-31.00	2.38E-21	1.11E-19	38.92
TSC2D3	Xq22.3	8.12	-3.14	-41.71	2.76E-22	3.28E-20	41.22	-1.66	-30.21	2.77E-19	1.48E-17	34.31	-2.19	-35.63	8.17E-21	3.29E-19	37.81
AXUD1	3p22.2	9.94	-3.15	-31.93	1.16E-21	1.08E-19	39.65	-1.03	-14.27	1.85E-13	1.57E-12	20.41	-2.19	-24.26	8.58E-19	1.87E-17	32.93
H3F3B	17q25.1	12.50	-3.34	-36.69	8.64E-24	2.32E-21	44.53	-1.42	-21.39	6.11E-18	1.93E-16	30.93	-4.40	-59.17	4.41E-29	6.35E-26	56.59
MTSS1	8q24.13	10.73	-3.35	-50.74	2.25E-27	3.27E-24	52.68	-1.47	-30.40	9.96E-22	1.44E-19	39.78	-3.00	-55.51	2.26E-28	1.94E-25	55.01
HBA2;HBA1	16p13.3	10.74	-3.37	-32.49	7.64E-22	7.54E-20	40.08	-2.63	-31.72	1.37E-21	1.82E-19	39.51	-1.68	-19.90	9.29E-17	1.21E-15	28.04
PELI1	2p14	12.50	-3.41	-31.91	2.93E-22	3.48E-20	40.99	-1.36	-17.41	8.87E-16	1.36E-14	25.83	-2.61	-29.93	1.46E-21	7.49E-20	39.30
FOS	14q24.3	10.57	-3.56	-53.02	7.32E-28	2.32E-24	53.76	-5.60	-114.10	2.07E-36	3.28E-32	70.61	-5.04	-91.87	5.47E-34	8.66E-30	67.12
CXCR4	2q21.3	11.74	-3.58	-42.79	9.25E-25	3.85E-22	46.78	-1.25	-18.67	4.11E-16	6.95E-15	26.74	-2.10	-30.70	3.01E-21	1.36E-19	38.63
COPEB	10p15.1	11.20	-3.66	-54.48	1.70E-26	1.49E-23	50.67	-3.15	-64.18	3.53E-28	9.31E-25	54.17	-3.54	-57.70	4.38E-27	2.66E-24	52.00
CD69	12p13.31	11.36	-3.77	-33.26	4.30E-22	4.70E-20	40.65	-3.44	-41.48	1.98E-24	1.08E-21	46.01	-4.36	-47.09	8.88E-26	2.81E-23	49.14
RGS1	1q31.2	9.77	-3.98	-58.73	5.35E-29	4.23E-25	56.26	-3.52	-71.22	3.80E-31	3.00E-27	60.67	-4.67	-84.42	4.81E-33	3.81E-29	65.17
FOSB	19q13.32	10.77	-4.47	-32.57	3.00E-21	2.30E-19	38.75	-6.06	-60.44	1.46E-27	2.32E-24	52.87	-6.36	-46.31	7.85E-25	1.53E-22	46.92
HBD;HBB	11p15.4	11.53	-4.54	-30.69	7.84E-22	7.54E-20	40.00	-2.59	-23.99	3.65E-19	1.83E-17	33.80	-2.25	-18.60	1.82E-16	2.19E-15	27.29
DUSP1	5q35.1	11.81	-4.65	-39.69	1.18E-24	4.73E-22	46.52	-4.62	-54.10	4.37E-28	9.87E-25	54.17	-5.30	-55.44	2.33E-28	1.94E-25	54.96
RGS2	1q31.2	11.54	-4.76	-49.62	3.99E-27	4.85E-24	52.12	-3.71	-52.88	7.84E-28	1.38E-24	53.61	-3.76	-47.98	9.37E-27	5.11E-24	51.34
TSC2D3	Xq22.3	12.20	-4.98	-60.70	1.32E-27	3.09E-24	53.09	-2.98	-45.38	1.27E-24	7.71E-22	46.37	-3.53	-47.19	5.05E-25	1.10E-22	47.40