

Supplementary Information

Figure Legends

Supplementary Figure 1. Pathologist scoring of BCLW and BCL2 IHC. A and B) Immunohistochemistry (IHC) for BCLW and BCL2 was performed on human tissue samples. A) Human kidney served as a control for BCLW IHC, as it shows no staining in the glomeruli (arrow) and positive staining within the tubules. B) Representative images of B-cell lymphoma patient samples for each score given by the pathologist to illustrate the scoring used for all lymphoma samples subjected to IHC.

Supplementary Figure 2. *BCLW* expression does not correlate with *BCL2* in GCB DLBCL and does not predict survival of ABC DLBCL. A) Gene expression-profiling data showing association between *BCLW* and *BCL2* mRNA in GCB DLBCL stratified based on the lowest ($n=148$) and highest ($n=148$) quartiles of *BCL2* expression. The lines represent the mean \pm SEM and the symbols represent individual samples. B) Patient samples of ABC DLBCL with low *BCL2* from the GSE10846 and GSE31312 data sets were each stratified based on the median expression of *BCLW* into two groups, *BCLW* low and *BCLW* high. Kaplan-Meier overall survival analyses were performed and compared using log-rank tests.

Supplementary Figure 3. *BCLW* and *BCL2* are overexpressed at a similar frequency in follicular lymphoma. A) Gene expression-profiling data showing the association between *BCLW* and *BCL2* mRNA in follicular lymphoma (FL) stratified based on the median expression of *BCL2* (left) into low ($n=98$) and high ($n=98$), or based on the lowest ($n=49$) and highest

($n=49$) quartiles of *BCL2* expression (right). The lines represent the mean \pm SEM and the symbols represent individual samples. B and C) qRT-PCR analysis for *BCLW* and *BCL2*, in triplicate, from FL patient samples ($n=30$) compared with normal lymph nodes ($n=12$) in B, or stratified based on grade (low, $n=19$; high, $n=11$) then plotted from lowest to highest *BCLW* expression for both grades in C. mRNA levels were compared to β -*ACTIN* and presented as $2^{-\Delta\Delta CT}$. Error bars are mean \pm SEM.

Supplementary Figure 4. Marginal zone lymphomas frequently overexpress BCLW. A)

Gene expression-profiling data showing the association between *BCLW* and *BCL2* mRNA in marginal zone lymphoma (MZL) stratified based on the median expression of *BCL2* into two groups, low ($n=74$) and high ($n=74$). The lines represent the mean \pm SEM and the symbols represent individual samples. $*P=8.73 \times 10^{-6}$; two-tailed *t*-test. B) qRT-PCR analysis for *BCLW* and *BCL2*, in triplicate, from nodal MZL patient samples ($n=10$) compared with normal lymph nodes ($n=12$). mRNA levels were compared to β -*ACTIN* and presented as $2^{-\Delta\Delta CT}$. Error bars are mean \pm SEM.

Supplementary Figure 5. BCLW is overexpressed in mantle cell lymphoma. A)

Gene expression-profiling data showing the association between *BCLW* and *BCL2* mRNA in mantle cell lymphoma (MCL) stratified based on the median expression of *BCL2* (left) into low ($n=98$) and high ($n=98$), or based on the lowest ($n=49$) and highest ($n=49$) quartiles of *BCL2* expression (right). The lines represent the mean \pm SEM and the symbols represent individual samples. B) qRT-PCR analysis for *BCLW* and *BCL2*, in triplicate, from MCL patient samples ($n=14$)

compared with normal lymph nodes ($n=12$). mRNA levels were compared to β -*ACTIN* and presented as $2^{-\Delta\Delta CT}$. Error bars are mean \pm SEM.

Supplementary Figure 6. Hodgkin lymphoma preferentially selects for BCLW overexpression. A) Gene expression-profiling data showing the association between *BCLW* and *BCL2* mRNA in Hodgkin lymphoma (HL) stratified based on the median expression of *BCL2* into two groups, low ($n=90$) and high ($n=90$). The lines represent the mean \pm SEM and the symbols represent individual samples. $*P=1.33\times 10^{-4}$; two-tailed *t*-test. B) qRT-PCR analysis for *BCLW* and *BCL2*, in triplicate, from HL patient samples ($n=26$ total; nodular sclerosis $n=16$; mixed cellularity $n=10$) compared with normal lymph nodes ($n=12$). mRNA levels were compared to β -*ACTIN* and presented as $2^{-\Delta\Delta CT}$. Error bars are mean \pm SEM. C) Box and whisker plots of data shown in B. The box represents the 25th and 75th percentiles, the line indicates the median, the circle indicates the mean, and the whiskers are the maximum and minimum, $*P=9.00\times 10^{-3}$; $**P=3.40\times 10^{-3}$; two-tailed *t*-tests.

Supplementary Figure 7. Expression of anti-apoptotic BCL2 family members in follicular lymphoma. A-C) qRT-PCR analysis for *BCLX*, *MCL1*, and *AI* mRNA, in triplicate, from follicular lymphoma (FL) patient samples ($n=30$) compared with normal lymph nodes ($n=12$). mRNA levels were compared to β -*ACTIN* and presented as $2^{-\Delta\Delta CT}$. Error bars are mean \pm SEM. Expression for each sample is shown on the left and averaged on the right. For the box and whisker plots, the box represents the 25th and 75th percentiles, the line indicates the median, the circle indicates the mean, and the whiskers are the maximum and minimum. For A, $*P=3.77\times 10^{-4}$; two-tailed *t*-test.

Supplementary Figure 8. Validation of *BCLW* overexpression in DLBCL. To illustrate that the top quartile of *BCLW* expression is biologically meaningful and contains samples where *BCLW* is highly overexpressed, the fold-change of *BCL2*, *BCLX*, *MCL1*, and *AI* expression was plotted and compared to that of the top 25% of *BCLW* expression in DLBCL. The vertical dashed line marks 1.5 fold, and samples that fall below 1.5-fold are considered to have a low level of expression.

Supplementary Table 1. Microarray gene expression profiling datasets.

GEO Acc. No.	Normal samples (<i>n</i>)	Tumor samples (<i>n</i>)
GSE12195	Centroblasts (5) Centrocytes (5) Memory B-cells (5)	DLBCL ^A (73) FL ^B (38)
GSE12453	Centroblasts (5) Centrocytes (5) Memory B-cells (5)	BL ^C (5) DLBCL (11) FL (5) HL ^D (17)
GSE12366	Germinal Center B-cells (3) Memory B-cells (3)	
GSE55267	Centroblasts* (1) Centrocytes* (1)	FL (63)
GSE65135	CD19+ B-cells from PB ^E (5)	DLBCL (2) FL (14) MZL (2)
GSE26725	CD19+ B-cells from PB (5)	
GSE500006	CD19+ B-cells from PB (32)	
GSE39133	Germinal center B-cells (5)	HL (29)
GSE39132	Centroblasts (5)	
GSE25638	CD19+ B-cells from PB (2) Centroblasts (2) Memory B-cells (2)	DLBCL (26) FL (15) MZL ^F (43)
GSE56314	Centroblasts (7) Centrocytes (7) Memory B-cells (7)	
GSE26673		BL (16)
GSE64085		BL [†] (11)
Dave et. al., 2006 [#]		DLBCL (74) BL (25)
GSE34171		DLBCL (91)
GSE74266		DLBCL (28)
GSE64555		DLBCL (40)
GSE23501		DLBCL (69)
GSE19246		DLBCL [§] (59)
GSE17372		DLBCL (13)
GSE10846		DLBCL [¶] (417)
GSE31312		DLBCL (498)
GSE56313		DLBCL (89)
GSE81183		FL (42)
GSE16455		FL (7) MZL (4) MCL ^G (22)
GSE16024		FL (7) MCL (7)
GSE35426		FL (5) MZL (14) MCL (5)
GSE35348		MZL (27)
GSE35082		MZL (9)

GSE25550	MZL (14)
GSE13314	MZI (35)
GSE17920	HL (130)
GSE14879	HL (4)
GSE19243	MCL (5)
GSE36000	MCL (38)
GSE21452	MCL (64)
GSE70910	MCL (55)

^ADLBCL, diffuse large B-cell lymphoma; ^BFL, follicular lymphoma; ^CBL, Burkitt lymphoma; ^DHL, Hodgkin lymphoma; ^EPB, peripheral blood; ^FMZL, marginal zone lymphoma; ^GMCL, mantle cell lymphoma.

*Two replicate samples were averaged; [§]fresh/frozen tissue samples amplified by Eberwine oligo-dT method were analyzed; [£]sample GSM1564138 was excluded from the analyses as it was not used in the original publication; [#]see supplementary ref.; [¥]3 CD19- samples with unknown disease status were excluded

Reference

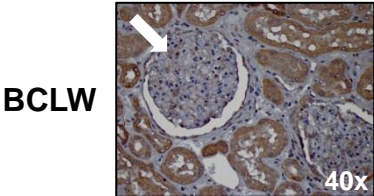
Dave SS, Fu K, Wright GW, Lam LT, Kluin P, Boerma EJ, et al. Molecular diagnosis of Burkitt's lymphoma. *N Engl J Med* 2006;354(23):2431-42.

Supplementary Table 2. Significantly inversely correlated gene pairs.

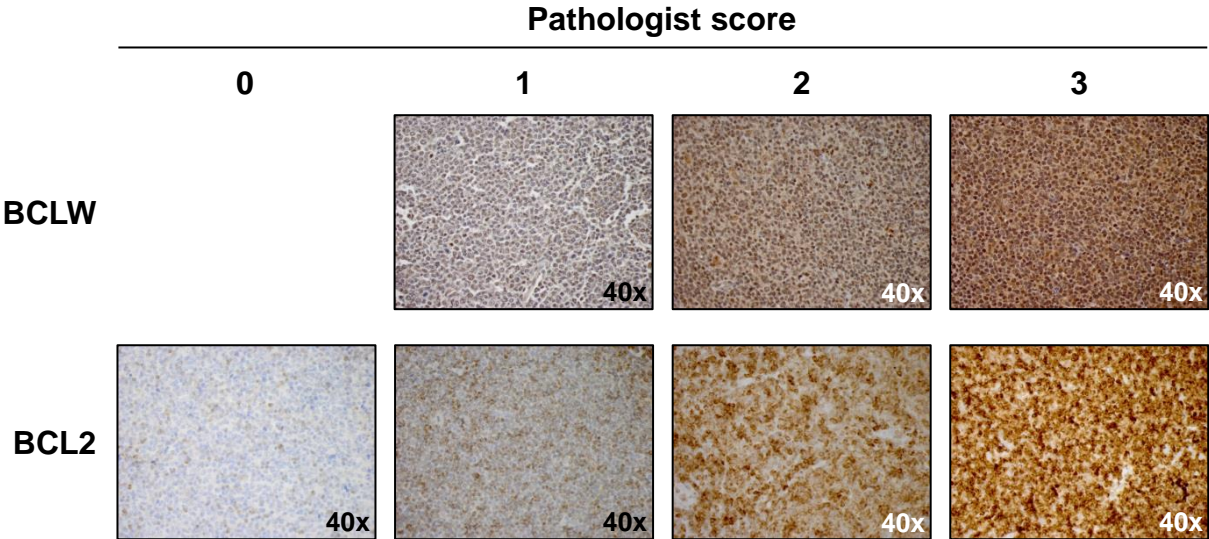
Genes compared	Lymphoma type	Pearson correlation			Spearman-rank correlation		
		coefficient	95% CI	P-value	coefficient	95% CI	P-value
<i>BCLW-BCL2</i>	DLBCL ABC	-0.201	-0.283 - -0.117	3.70E-06	-0.199	-0.281 - -0.115	4.90E-06
<i>BCLW-BCL2</i>	MZL	-0.384	-0.514 - -0.238	1.40E-06	-0.379	-0.509 - -0.231	2.50E-06
<i>BCLW-BCL2</i>	Hodgkin	-0.349	-0.471 - -0.213	1.61E-06	-0.316	-0.442 - -0.178	1.70E-05
<i>BCL2-MCL1</i>	MZL	-0.220	-0.368 - -0.061	7.28E-03	-0.243	-0.389 - -0.085	2.97E-03
<i>BCL2-BCLX</i>	MZL	-0.207	-0.357 - -0.048	0.011	-0.183	-0.335 - -0.023	0.026
<i>BCLW-AI</i>	Hodgkin	-0.294	-0.422 - -0.154	6.17E-05	-0.302	-0.429 - -0.163	4.19E-05

CI – Confidence interval

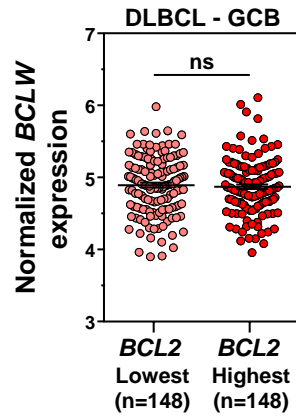
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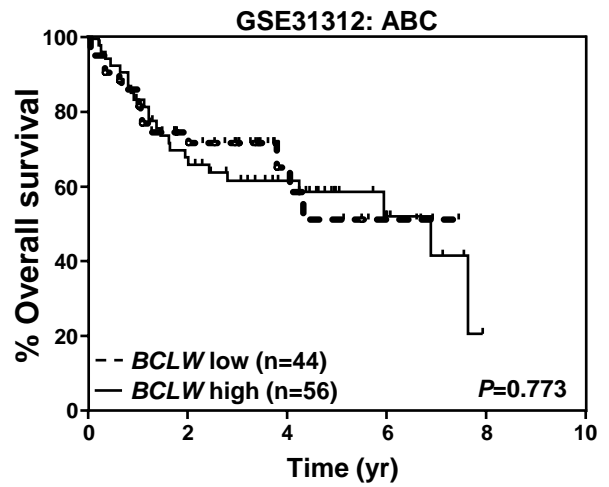
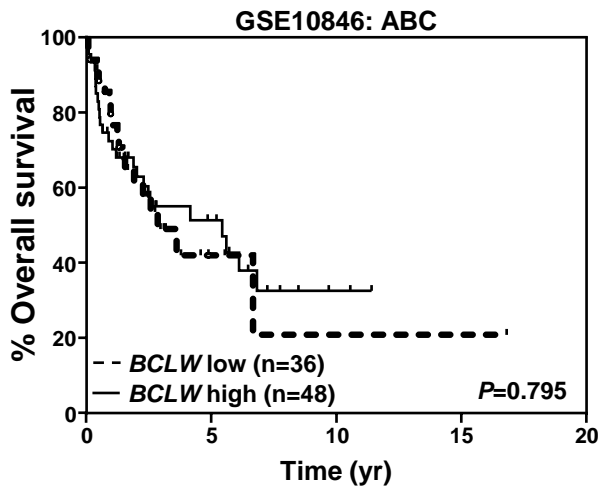
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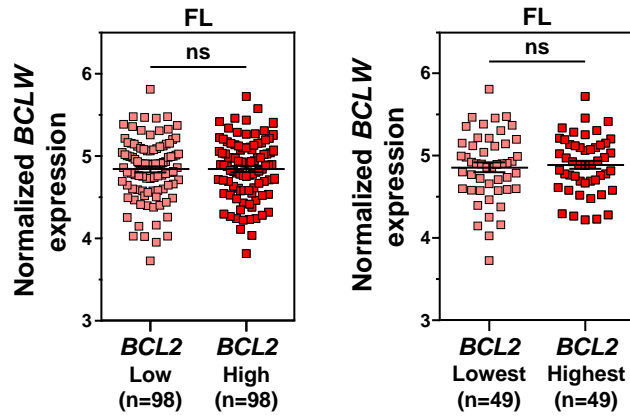
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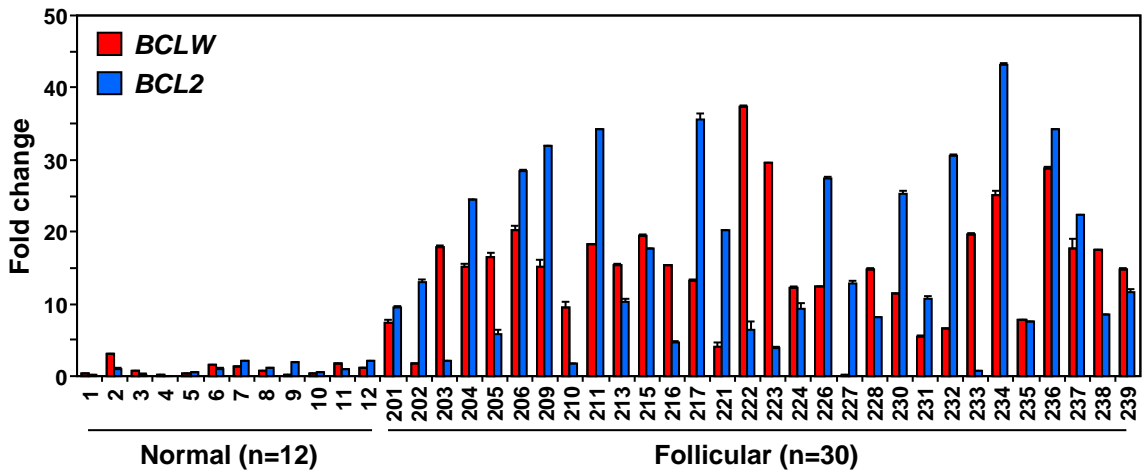
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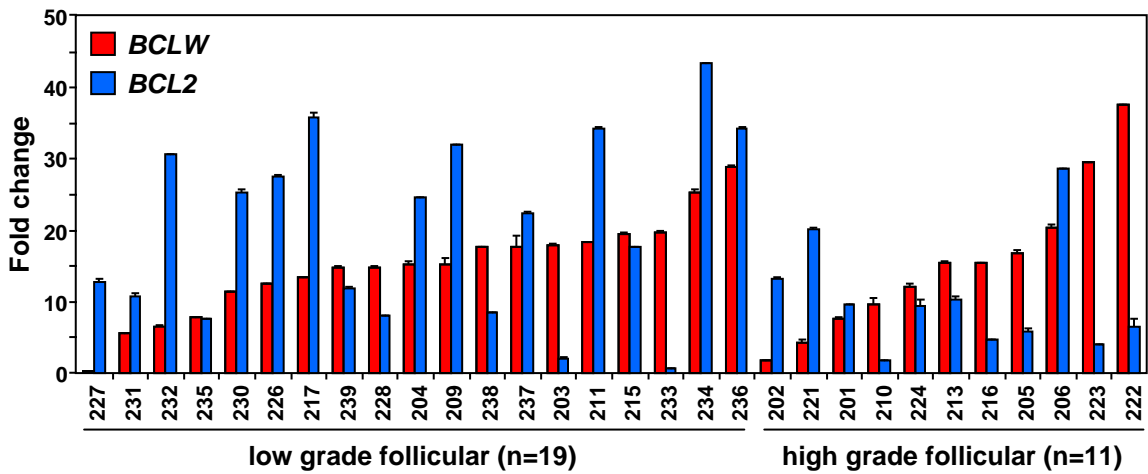
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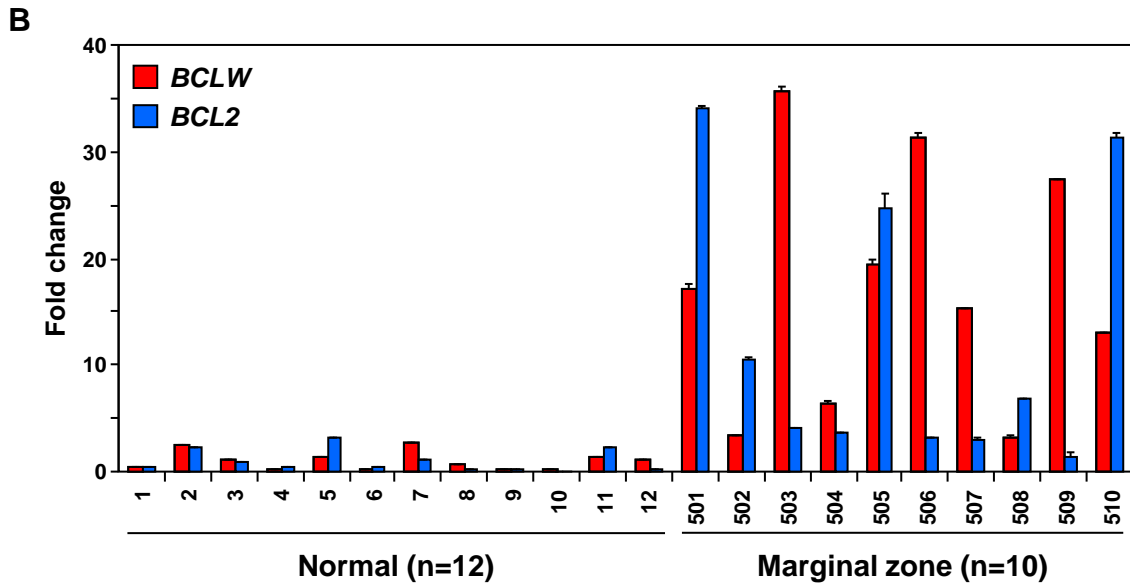
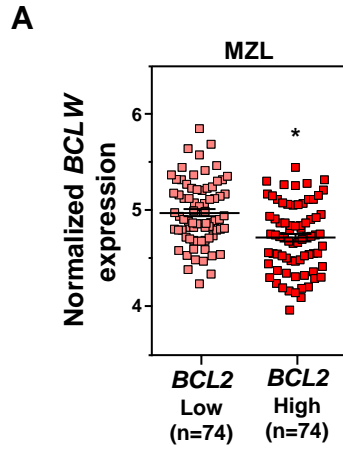


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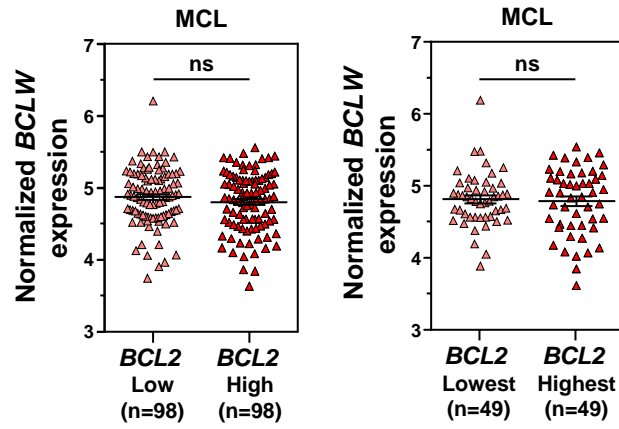


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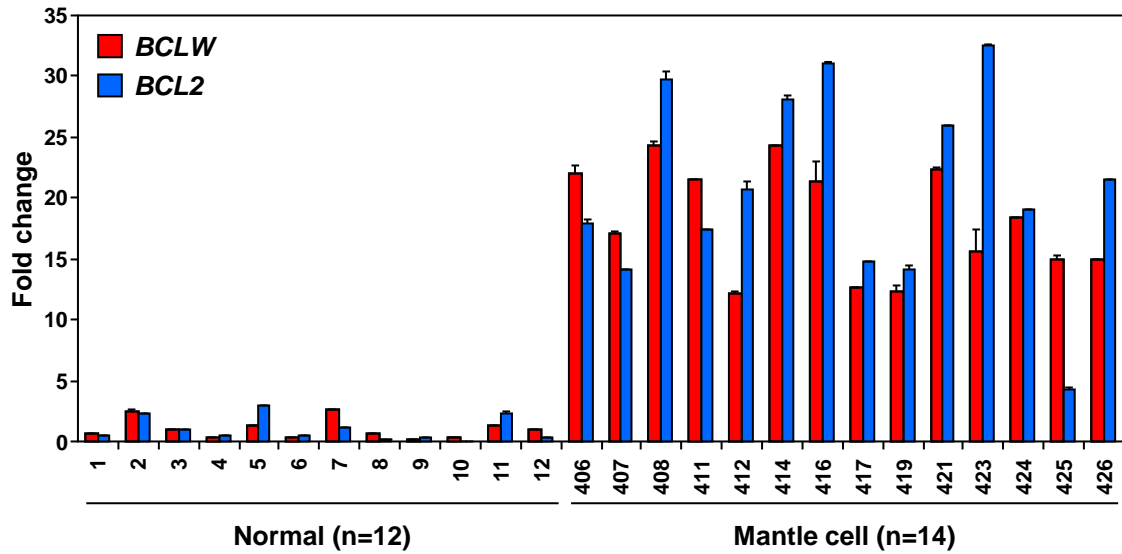


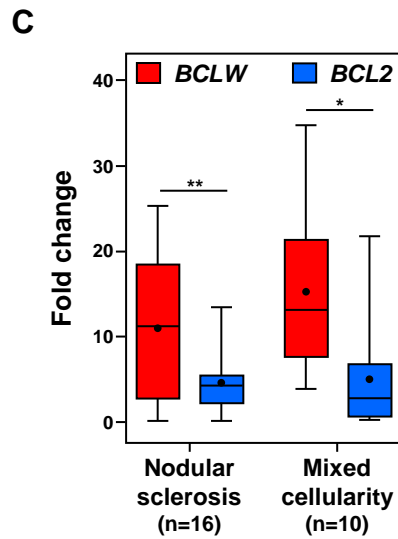
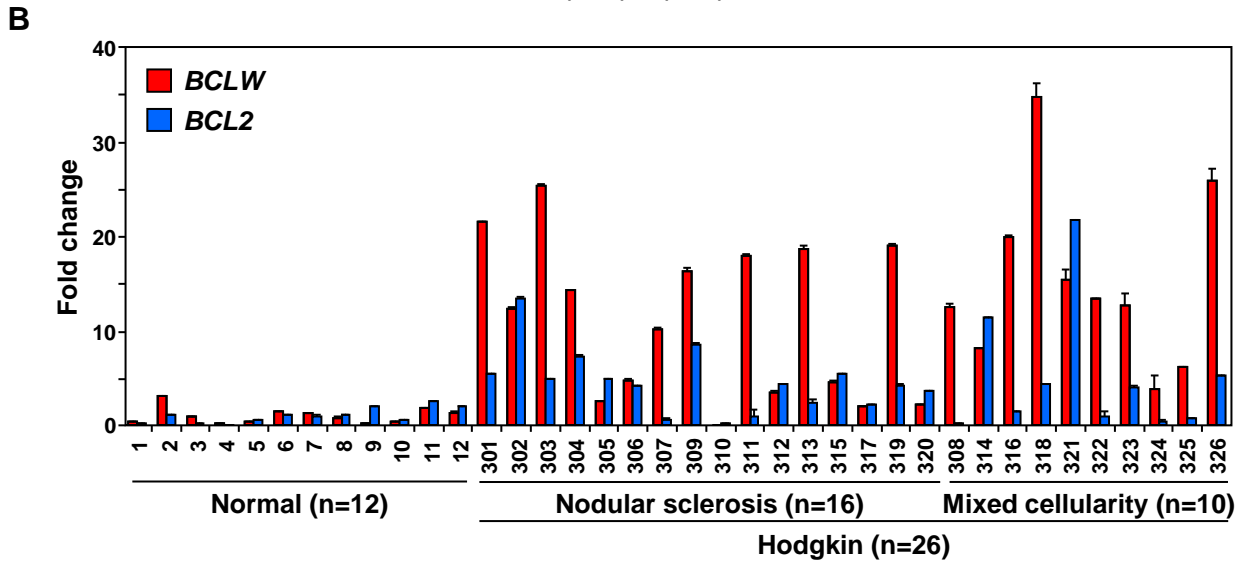
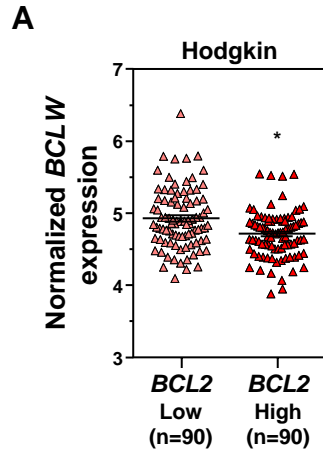


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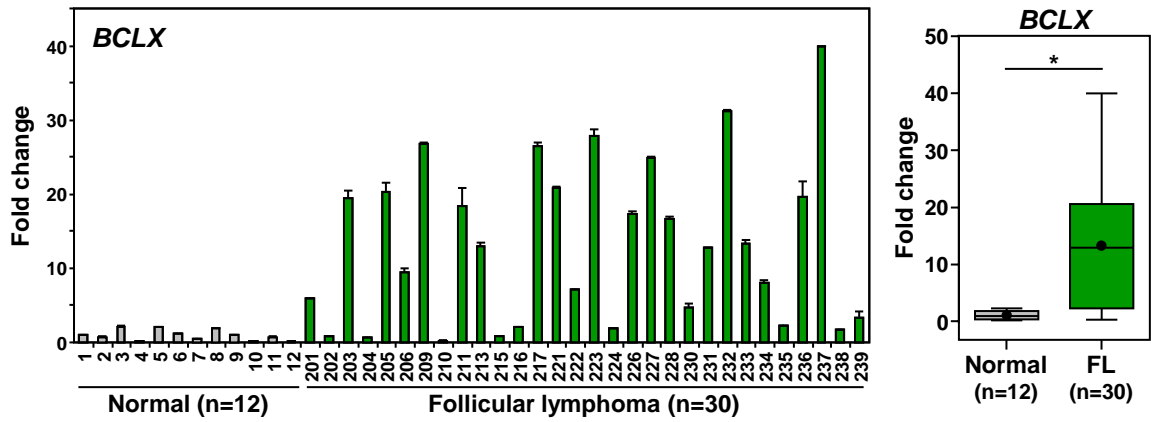


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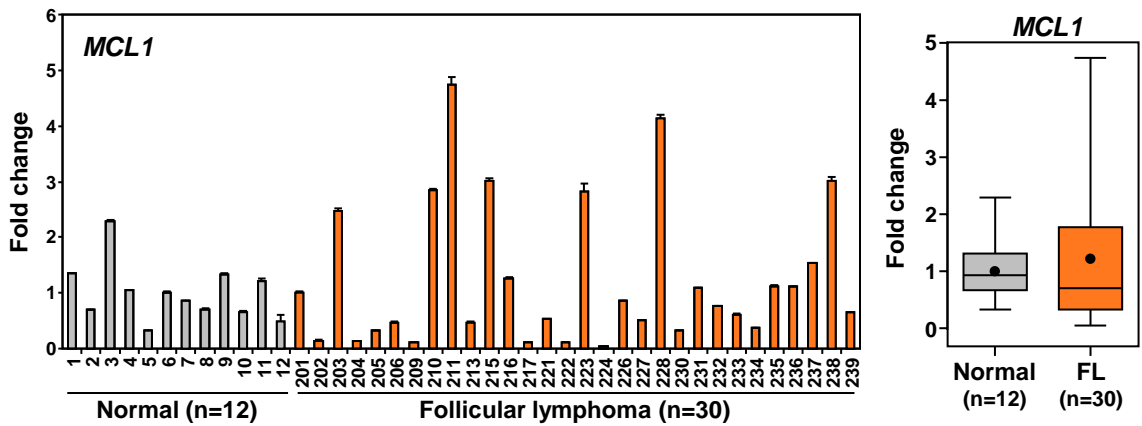




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