# Global miRNA profiling reveals key molecules that contribute to different chronic lymphocytic leukemia incidences in Asian and Western populations

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# Supplementary Materials for "Global microRNA profiling reveals key molecules that contribute to different CLL incidences in Asian and Western populations

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## Supplementary Methods

### MicroRNA microarray assay

The Affymetrix GeneChip microRNA 3.0 Array (Affymetrix, Santa Clara, CA, USA) analyses were performed using the standard procedures recommended by the manufacturer. This microRNA array contains probe sets for 5607 human miRNAs. The RNA labeling, microRNA array hybridization, scanning, and quantification were performed according to the Affeymetrix GeneChip protocols. The CEL-files of the raw data were obtained using the Affymetrix GeneChip Command Console Software, and the data was pre-processed by Affymetrix Expression Console software 1.2 using the default analysis settings, and Robust Multi-array Average (RMA) method was used for normalization. miRNAs with values above the cutoff of 10 in all samples were chosen for data analysis. The expression values were generated and subsequently normalized using the Affymetrix Gene Chip Command Console (AGCC) Software. In the microRNA profiling, probes with detection rate less than 80% across the subjects were removed to minimize false positive results and improve data quality.

### Quantitative real-time PCR

Total RNA was extracted using the TRIzol reagent (Invitrogen, USA). Conversion of mRNA to cDNA was accomplished using the Reverse Transcription System Kit (Promega, Madison, WI, USA) according to the manufacturer's instructions. cDNA was used for quantitative real-time PCR using the Bio-Rad CFX96 real-time PCR detection system (Bio-Rad Laboratories, Richmond, CA, USA), using specific primers in SYBR green reactions to determine representative mRNA levels. The following primers were used: TGR-5, F: 5'-CCCAGGCTATCTTCCCAGC-3', R: 5'-GCCAGGACTGAGAGGAGCA-3'; ASB7, F: 5'-CCGCTTCAGCTCGCCATTAT-3', R: 5'-GAATGTCGATGTTGGCATTGTG-3'; CTDSPL2, F: 5'-CAACTAATGGAGCAGCTTACTCA-3', R: 5'-CGCGTGGGCTGATGA ATAAC-3'; FBX032, F: 5'-TTCAAACTTGTCCGATGTTACCC-3', R: 5'-CCAGGAAA GGATGTGACAGTGT-3'; JOSD1, F: 5'-GGGATACGCTGCAAGAGATTT-3', R: 5'-CCATGACGTTA GTGAGGGCA-3'; MED1, F: 5'-GAAGTGTTGGCTATCTCACA CC-3', R: 5'-TGTCATCCAGT AGGTCAGAAGG-3'. β-actin was used as a control. The All-in-One™ microRNA qPCR Kit (GeneCopoeia, Guangzhou, China) was used for microRNA expression analysis of miR-4485, miR-138, miR-181c, miR-181d, miR181a and miR-363.

**Target gene prediction**

Target genes of miRNAs were predicted using miRWalk (http://mirwalk.umm.uni-heidelberg.de), TargetScan (https://www.targetscan.org), miRDB (https://mirdb.org), and miRPathDB (https://mpd.bioinf.uni-sb.de); A score of ≥ 0.95 was used as the cut-off criterion in the prediction analysis to select candidate target genes for further analysis.

**Establishment of stable cell lines with miR-4485 low or high expression**

Lentiviral miArrest$^{TM}$ microRNA inhibitor vector for microRNA-4485 containing the pEZX-AM03 vector, a mCherry reporter gene, a hygromycin selectable marker, and a H1 promoter (Cat. # HmiR-AN2125-AM03, Genecopoeia, China) and miExpress$^{TM}$ microRNA Expression Precursor for microRNA-4485 containing the pEZX-MR03 vector, an eGFP reporter gene, a puromycin selectable marker and a CMV promoter (Cat. # HmiR1168-MR03, Genecopoeia, China) were transduced into proper host cells. Polybrene (8 mg/ml, Sigma-Aldrich) was used in lentiviral particle infection of cells. After 24 hours incubation at 37℃, medium was replaced with fresh medium to exclude polybrene. The transduced cells were then selected with puromycin (Invivogen, San Diego, CA, USA) or hygromycin (Invitrogen, Carlsbad, CA, USA) for 2-3 weeks to obtain cells with stable overexpression or low expression of miR-4485. The respective vectors containing scrambled microRNA sequences were used in parallel experiments as controls.

**Transfection of microRNA oligonucleotides**

MicrON miR-4485-3p mimic (dsRNA oligonucleotides) (Cat. # miR10019019), micrOFF miR-4485 inhibitor (single-stranded oligonucleotides) (Cat. # miR2160621085914), micrON mimic negative control (Cat. # miR1N0000001-1-5) and micrOFF inhibitor negative control (Cat. # miR2N0000002-1-5) were acquired from RiboBio Co., Ltd (Guangzhou, China). Their sequences of oligonucleotides are as follows: micrONTM miR-4485 mimic, 5'-UAACGGCCGCGGUACCCUAA-3', 3'-AUUGCCGGCGCCAUGGGAUU-5'; micrOFFTM miR-4485 inhibitors, 5'-UUAGGGUACCGCGGCCGUUA-3'. The microRNA mimic and microRNA inhibitor were transfected into cells using a riboFECT CP transfection kit (Cat. C10511-05, RiboBio Co., Ltd). Transfection efficiency was confirmed by fluorescent probe co-transfected.

**Dual-luciferase reporter assay**

The 3′UTR fragments of TGR5 (also known as GPBAR1) were inserted into the luciferase reporter vector pmiR-RB-REPORT™ (RIBOBIO, Guangzhou, China) between the XhoI and NotI restriction sites. TGR5 3′UTR mutant (Figure 4D) was also constructed and confirmed by sequencing. For the luciferase reporter assay, HEK293T cells were plated at a density of $1.5x10^4$ cells/well in 96-well plates. After 24 hours, cells were transiently co-transfected with 10 μL of miR-4485-3p mimic or Non-target Control and 15 μL of wild-type or mutated TGR5 luciferase reporter vector, using Lipofectamine™ 2000 transfection reagent. After 48 hours, luciferase activity was measured using Dual-Glo® Luciferase Assay System (Promega, Fitchburg, USA). The relative luciferase activity was calculated as the ratio of renilla/firefly RLU normalized by the respective control reporter construct. Statistical significance was calculated by unpaired two-tailed t-test, using $P < 0.05$ as the value for statistical significance.

**Establishment of stable cell lines with TGR5-high or -low expression**

TGR5 overexpression plasmid (Cat. #PPL01052-2a) and shRNA-mediated knockdown plasmids (Cat. # PPL01052-3; Sequence #1: CGTCTACTTGGCTCCCAACTT; Sequence #2: CCTCATCATCACCGCGAACCT; #3: GCATTGCCTACCACCCAAGCA; Negative control: GTTCTCCGAACGTGTCACGTT) were purchased from GenePPL Technology Co, Ltd (Nanjing, China). Lentiviral particles containing the TGR5 plasmids were produced in 293T packaging cells. For viral infection, cells were seeded into 6-well plates and grown to 70%–80% confluency, and the culture medium was replaced by fresh RPMI-1640 medium containing the desired lentiviral particles. After 72 hours, the medium was replaced with normal culture medium. Cells with stable overexpression or knockdown of TGR5 were selected with 40 μg/mL G418 (TGR5 overexpression) or 2 μg/mL puromycin (TGR5-shRNA knockdown) for 3 days.

**Cell proliferation assay**

Cell proliferation assay was performed by plating the indicated number of cells ($1\times10^4$ cells/well for 12-well plates; $1\times10^5$ cells/well for 6-well plates) in their respective culture media, and cell numbers were directly counted every 24 hours using an automated cell counter (Cellometer AutoT4, Nexcelom, USA). Each data point represents the mean ± SD from three independent experiments.

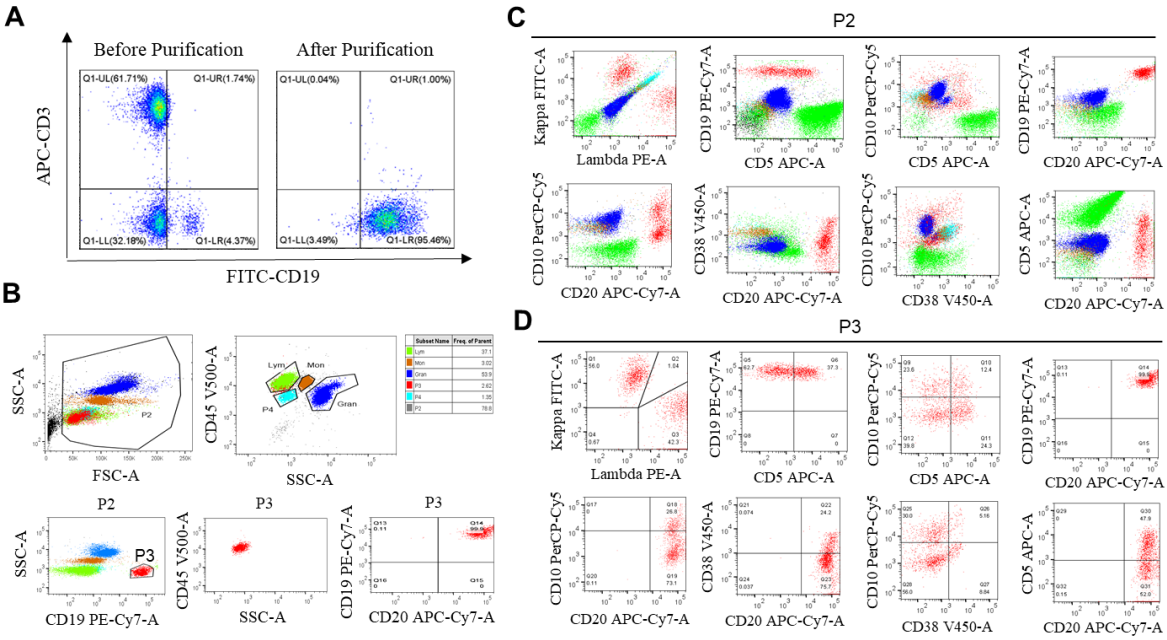
**Western blot analysis**

Cell samples were washed with ice-cold PBS and lysed in lysis buffer (containing RIPA and a cocktail of phosphatase inhibitors and protease inhibitors) for 15 minutes on ice. Cell debris was removed by centrifugation (12,000x g) for 15 minutes at 4°C. Protein lysates were analyzed by standard SDS-PAGE, followed by transferring to a nitrocellulose membrane. Proteins of interest were revealed by Western blotting using specific antibodies. Detail information on antibodies used in the Western blot is provided in *Online supplementary Table S5.* The ERK1/2 inhibitor SCH772984 (Cat. # HY-50846) and TGR5 agonist (Cat. # HY-14229) were purchased from MedChemExpress (Shanghai, China).

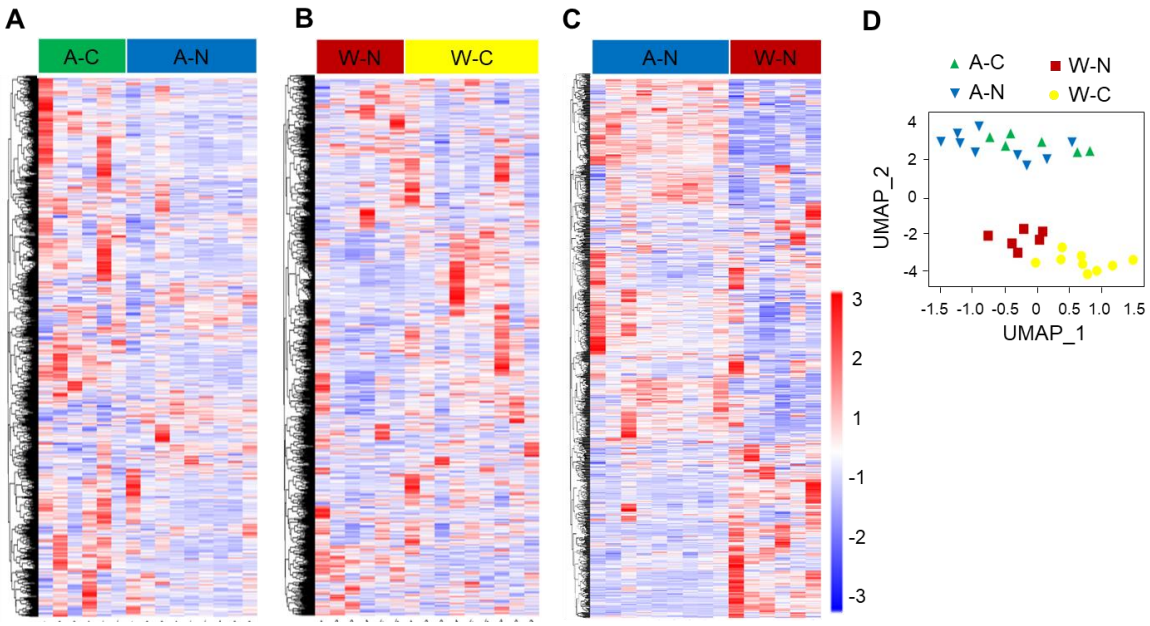
**Data analysis and statistical tests**

T-SNE (t-distribution stochastic neighbor embedding), a nonlinear dimensionality reduction algorithm with the ability of mapping data in the high-dimensional space to low-dimensional space and retain the local characteristics of the data set, and UMAP (uniform manifold approximation and projection) capable of preserving both local and global structures of the data, were used for microRNA data dimensionality reduction and unsupervised clustering visualization. Multivariate analyses including unsupervised principal component analysis (PCA) and supervised orthogonal projections to latent structures discriminant analysis (OPLS-DA) were employed to assess the data discriminations of various groups. OPLS-DA enables the screen of differential variables by removing the unrelated differences. A 200-permutation test was performed to assess the statistical significance and prevent overfitting of the OPLS-DA model. Two-tailed Student *t*-test was used to determine potential difference between two groups, analysis of variance (ANOVA) with a Tukey's *post hoc* test was applied when more than two groups were compared. For microRNA expression data from small sample number, *p* values were adjusted using Benjamini-Hochberg false discovery rate (FDR) to calculate *q* values. A *p* or *q* value of less than 0.05 was considered statistically significant. Differentially expressed miRNAs were identified using the criteria of fold change (FC) > 2 or log2 FC>1 and a *q or p* value <0.05. The results of t-SNE and UMAP dimensionality reduction were visualized using Sangerbox 3.0 (http://www.sangerbox.com/tool). Multivariate analysis was performed using SIMCA-16 software (Umetrics AB, Umeå, Sweden). Statistical analyses were performed using SPSS 25.0 software (IBM Corp, Armonk, NY, USA) or Graph Pad Prism version 9.0 (GraphPad software Inc., CA, USA).

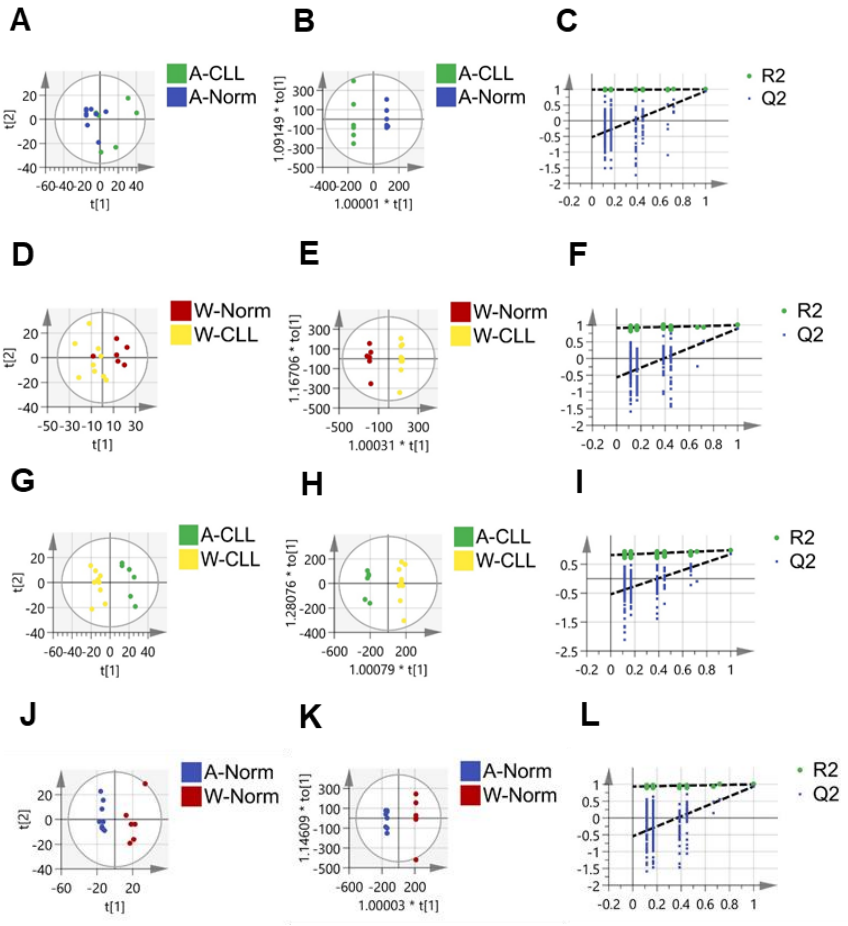
## Supplementary Figures



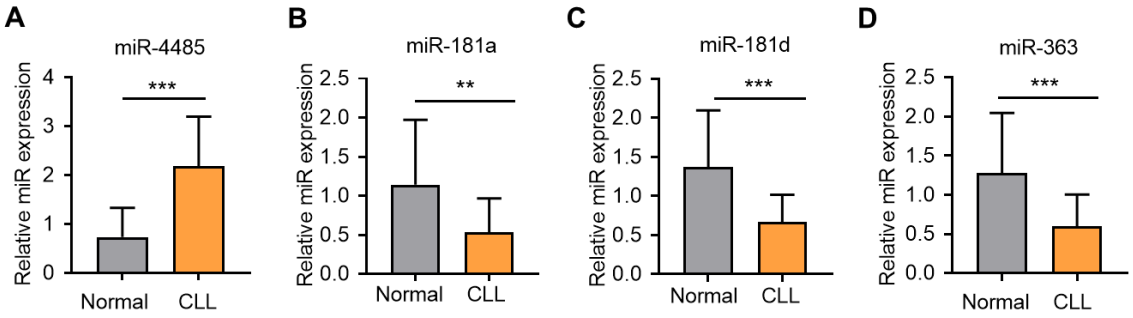

**Supplementary Figure 1. Flow cytometry analysis of B cell purity and monoclonal B cell lymphocytosis.** (A) B cells were analyzed based on their expression of CD19 and absence of T cell marker CD3. The purity of B cells was more than 95% after purification. (B-D) Multi-color flow cytometry analysis of normal peripheral blood to conform the absence for monoclonal B cell lymphocytosis. Whole blood cells were stained with various combinations of fluorochrome-conjugated antibodies as indicated, and specific sub-populations of cells were gated according to their light scattering and staining by specific antibodies. Cells were first gated on size and singularity to exclude dead cells. B-lymphocytes were gated based on CD19 expression. The presence or absence of monoclonal B cell lymphocytosis was evaluated by CD19, CD5, Kappa, and lambda immunophenotyping.



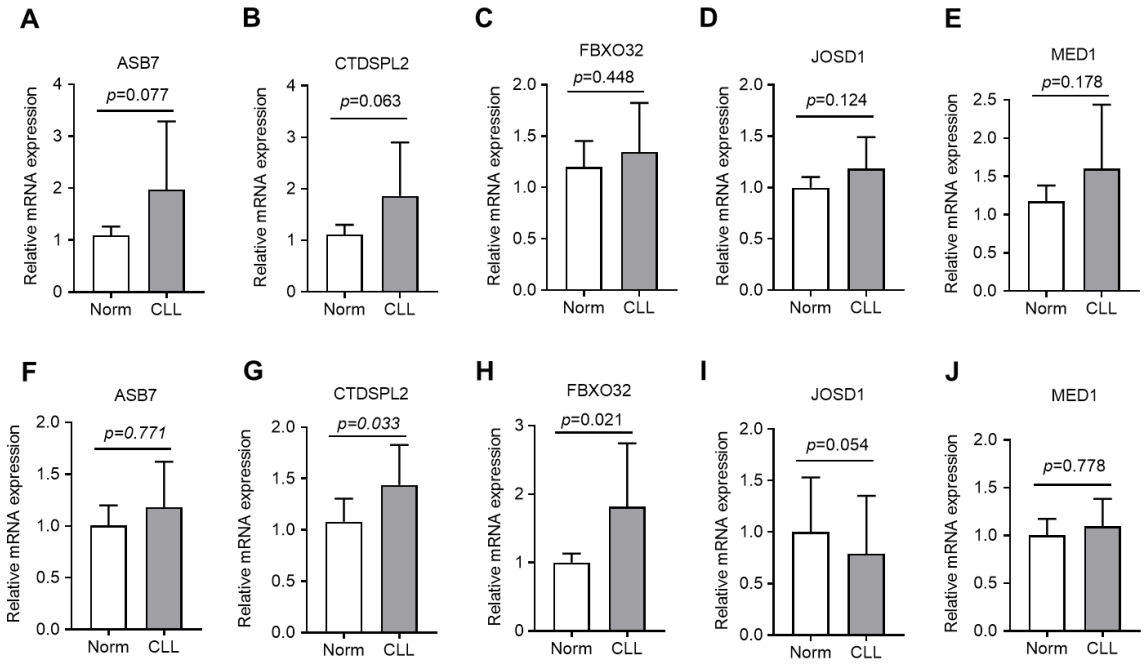

**Supplementary Figure 2. Unsupervised hierarchical clustering and UMAP visualization of microRNA expression.** (A, B, C) Unsupervised hierarchical clustering of microRNA expression by the indicated sample groups (A-C: Asian CLL, A-N: Asian normal, W-C: Western CLL, W-N: Western normal). Each row represents a microRNA, and each column represents an analyzed sample. The color code represents the relative intensity of the microRNA expression signal, with red indicating high expression and blue indicating low expression. (D) Uniform manifold approximation and projection (UMAP) plots depicting sample clustering according to microRNA expression profiles.



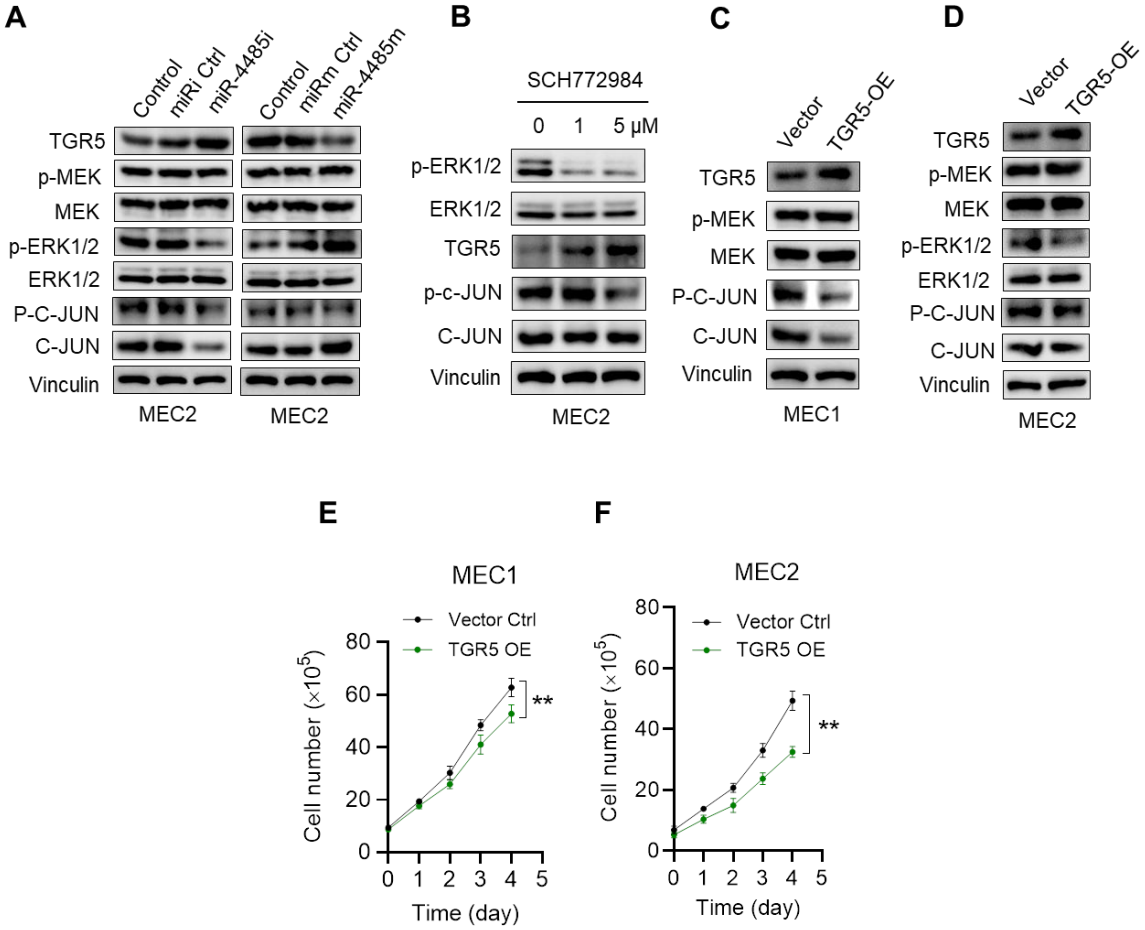

**Supplementary Figure 3. Analysis of microRNA expression in CLL cells and normal B lymphocytes from Asian and Western individuals.** (A) PCA score plots of microRNA expression in CLL cells vs normal B lymphocytes from Asian individuals. $R^2X$=0.551. (B) OPLS-DA score plots of microRNA expression in CLL cells vs normal B lymphocytes from Asian group. $R^2X$=0.877, $R^2Y$=1.000, $Q^2$=0.939. (C) Permutation test of OPLS-DA model in CLL cells vs normal B lymphocytes from Asian group, $Q^2$= -0.525. (D) PCA score plots of microRNA expression in CLL cells vs normal B lymphocytes from Western group. $R^2X$=0.550. (E) OPLS-DA score plots of microRNA expression in CLL cells vs normal B lymphocytes from Western group. $R^2X$=0.666, $R^2Y$=0.997, $Q^2$=0.894. (F) Permutation test of the OPLS-DA model in CLL cells vs normal B lymphocytes from Western group, $Q^2$= -0.566. (G) PCA score plots of microRNA expression in CLL cells from Asian and Western groups. $R^2X$=0.535. (H) OPLS-DA score plots of microRNA expression in CLL cells from Asian and Western groups. $R^2X$=0.500, $R^2Y$=0.987, $Q^2$=0.847. (I) Permutation test result of the OPLS-DA model in CLL cells from Asian and Western groups, $Q^2$= -0.539. (J) PCA score plots of microRNA expression in normal B lymphocytes from Asian and Western groups. $R^2X$=0.572. (K) OPLS-DA score plots of microRNA expression in normal B lymphocytes from Asian and Western groups. $R^2X$=0.832, $R^2Y$=0.999, $Q^2$=0.938. (L) Permutation test result of the OPLS-DA model in normal B lymphocytes from Asian and Western groups, $Q^2$= -0.545. PCA: Principal component analysis; OPLS-DA: Orthogonal projections to latent structures discriminant analysis; PT, permutation test.



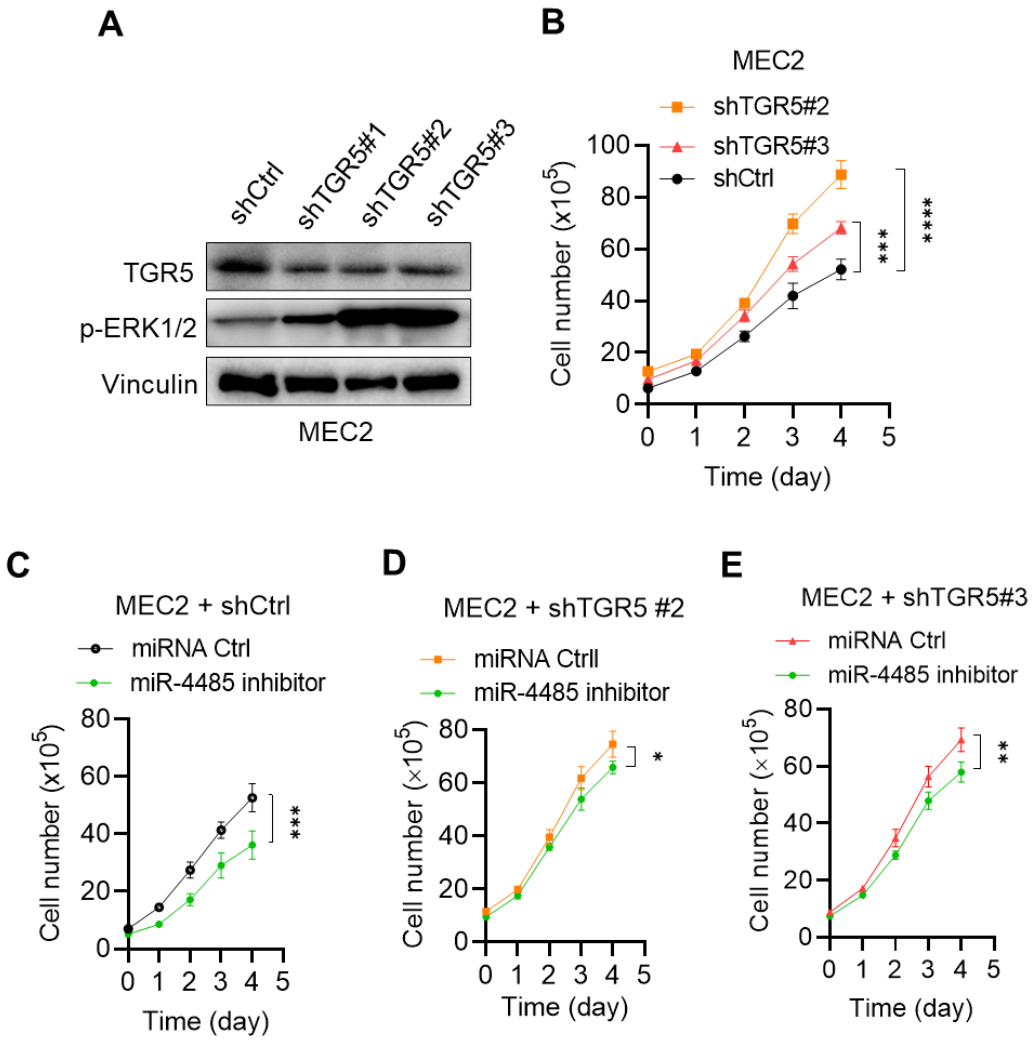

**Supplementary Figure 4**. Comparison of microRNA expression in normal B lymphocytes from healthy individuals and primary CLL cells from patients. Two microRNA expression datasets (GSE108901 and GSE66186) from Gene Expression Omnibus (GEO) database were used for analysis of expression of miR-4485 (A), miR181a (B), miR-181d (C), and miR-363 (D) in normal lymphocytes from healthy doners (n=20) and primary CLL cells from patients (n=34). To ensure comparability of the two datasets, the series matrix files were downloaded and merged with the inSilicoMerging R software package to adjust for batch effect, using the empirical Bayes methods (DOI: 10.1093/biostatistics/kxj037). Since GSE108901 dataset did not contain miR-181c and miR-138, these two miRNAs were not compared. **, $P<0.01$; ***, $P<0.001$.




**Supplementary Figure 5. Comparison of mRNA expression of *ASB7*, *CTDSPL2*, *FBXO32*, *JOSD1* and *MED1* genes in CLL cells and normal lymphocytes.** (A-E) Expression of the indicated genes in CLL cells from Asian patients (n=11) and normal lymphocytes from healthy Asian individuals (n=8). Gene expression was quantified by qRT-PCR. (F-J) Comparison of mRNA expression of the indicated genes in CLL cells from Western patients (n=45) and normal lymphocytes from healthy Western individuals (n=5). The genes expression data were from GEO database (GSE66117). None of the genes met the criteria of more than 2-fold change in expression (FC > 2) and a *P*-value of less than 0.05 to be considered significant.




**Supplementary Figure 6**. **Role of TGR5 in mediating miR-4485 effect on ERK1/2 and cell proliferation.** (A) MEC2 cells were transfected with miR-4485 mimic or inhibitor oligonucleotides in parallel with their respective oligo controls, and the expression of TGR5, phosphorylated MEK1/2, MEK1/2, ERK1/2, phosphorylated ERK1/2, phosphorylated-c-Jun and c-Jun was measured by Western blot analysis. (B) MEC2 cells were treated with or without the ERK inhibitor SCH772984 (1-5μM, 48 hours), and the indicated molecules were then analyzed by Western blot. (C) MEC1 cells were transduced with TGR5 expression vectors (TGR5-OE) or control vector, and cells were collected for Western blot analysis of indicated proteins. (D) MEC2 cells were transduced with TGR5 expression vectors (TGR5-OE) or the control vector, and the cells were collected for Western blot analysis of indicated proteins. miR-4485i: miR-4485 inhibitor; miRi Ctrl, microRNA inhibitor control; miR-4485m: miR-4485 mimic; miRm Ctrl: microRNA mimic control. (E, F) MEC1 and MEC2 cells were transfected with TGR5 overexpression vector (TGR5-OE) or the vector control, and cell numbers were counted every 24 hours.




**Supplementary Figure 7. Impact of TGR5 on ERK1/2 activation and cell proliferation in MEC2 cells.** (A) Effect of TGR5 knockdown by three shRNA (#1, #2, #3) on ERK1/2 phosphorylation in MEC2 cells, measured by Western blotting. (B) MEC2 cells were transfected with TGR5 knockdown vectors (shTGR5#2, shTGR5#3) or the negative control (shCtrl), and cell proliferation was measured by direct cell counting every 24 hours. (C-E) MEC2 cells transduced with empty vector or shTGR5 to stably knockdown TRG5. The cells with or without TGR5 knockdown were then transfected with miR-4485 inhibitor or the control oligos. Cell numbers were counted every 24 hours. Two-way ANOVA analysis was used to determine the significance of change in cell proliferation. Error bars indicate SD from three independent experiment; *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$; ****, $P < 0.0001$.

# Supplementary Tables

## Supplementary Table 1. Demographic information of CLL patients whose blood samples were used in this study

| Patient # | Race | Sex/Age | WBC ($x10^9$/L) | Treatment status | IGVH | Del (13q) | Del (17p) | Del (11q) | TP53 mut | Trisomy 12 | Trisomy 19 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | W | F/68 | 240.1 | Untreated | M | Yes | No | No | No | No | No |
| 2 | W | M/73 | 157.4 | Untreated | M | Yes | No | No | No | No | No |
| 3 | W | M/59 | 49.7 | Untreated | M | Yes | No | Yes | No | No | No |
| 4 | W | F/79 | 263.0 | Previously Treated | UM | Yes | No | No | Yes | No | No |
| 5 | W | M/81 | 293.0 | Untreated | M | Yes | No | No | No | No | No |
| 6 | W | F/78 | 107.5 | Untreated | M | Yes | No | No | No | No | No |
| 7 | W | M/55 | 38.9 | Untreated | UM | No | Yes | No | No | No | No |
| 8 | W | M/60 | 117.2 | Untreated | UM | No | No | No | No | Yes | Yes |
| 9 | W | M/74 | 109.3 | Untreated | UM | Yes | No | No | No | No | No |
| 10 | A | F/75 | 262.8 | Untreated | M | No | Yes | No | Yes | No | No |
| 11 | A | M/36 | 35.9 | Untreated | UM | Yes | Yes | No | Yes | No | No |
| 12 | A | M/65 | 56.2 | Untreated | UM | No | No | No | No | No | No |
| 13 | A | F/53 | 31.5 | Untreated | UM | No | Yes | No | No | No | No |
| 14 | A | M/70 | 45.0 | Untreated | M | No | No | No | No | Yes | No |
| 15 | A | M/70 | 200.2 | Untreated | UM | Yes | No | No | No | No | No |
| 16 | A | F/57 | 26.0 | Untreated | UM | Yes | No | No | No | No | No |
| 17 | A | F/78 | 12.3 | Previously treated | M | No | Yes | No | Yes | No | No |
| 18 | A | M/68 | 32.0 | Untreated | M | No | No | No | Yes | No | No |
| 19 | A | M/27 | 52.7 | Untreated | M | No | Yes | No | Yes | No | No |
| 20 | A | M/46 | 41.0 | Untreated | UM | No | No | No | No | No | No |
| 21 | A | M/71 | 37.1 | Untreated | M | No | No | No | Yes | No | No |
| 22 | A | M/60 | 34.18 | Untreated | M | No | No | No | Yes | No | No |
| 23 | A | M/52 | 99.84 | Untreated | M | No | Yes | No | Yes | No | No |
| 24 | A | M/50 | 56.81 | Untreated | UM | No | No | No | No | No | No |
| 25 | A | M/54 | 74.22 | Untreated | M | No | Yes | No | No | No | No |
| 26 | A | M/61 | 78.23 | Untreated | UM | No | Yes | No | Yes | No | No |
| 27 | A | M/64 | 104.46 | Untreated | M | No | Yes | No | No | No | No |
| 28 | A | M/70 | 164.29 | Untreated | UM | Yes | No | No | No | No | No |
| 29 | A | M/65 | 18.1 | Untreated | M | No | Yes | No | Yes | No | No |
| 30 | A | F/63 | 70.07 | Untreated | M | No | No | No | No | Yes | No |
| 31 | A | M/63 | 94.59 | Untreated | UM | Yes | No | No | Yes | No | No |

**Notes**: WBC, white blood cell counts at diagnosis; F, female; M, male; Treatment status shows if the patient was previously treated or untreated at the time of blood drawing; IGVH, immunoglobulin heavy-chain variable region; W, Western; A, Asian; M, mutated, UM, Unmutated.

**Supplementary Table 2. Differential microRNA expression in CLL cells and normal B lymphocytes from Asian individuals**

| Gene ID | Fold Change (CLL/Norm) | Asian samples- | | | | | | | | | | | | | | | q-value % |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | N1 | N2 | N3 | N4 | N5 | N6 | N7 | N8 | N9 | C1 | C2 | C3 | C4 | C5 | C6 | |
| miR-1295 | 14.24 | 3.6 | 2.6 | 2.3 | 3.9 | 6.2 | 1.4 | 4.4 | 8.4 | 3.3 | 212.8 | 74.3 | 102.1 | 3.5 | 110.3 | 27.6 | 0.00 |
| miR-4485 | 9.49 | 39.0 | 14.1 | 20.2 | 15.2 | 21.0 | 18.9 | 18.7 | 22.7 | 10.0 | 1524.8 | 239.4 | 440.4 | 67.9 | 19.9 | 146.1 | 0.00 |
| miR-4524-star | 9.49 | 1.3 | 1.7 | 1.5 | 1.5 | 1.9 | 1.4 | 1.4 | 2.0 | 1.6 | 1.5 | 35.8 | 24.8 | 23.4 | 13.3 | 26.8 | 0.00 |
| miR-34a | 3.97 | 9.8 | 17.9 | 30.5 | 13.6 | 21.4 | 16.7 | 18.1 | 15.9 | 16.9 | 100.6 | 8.4 | 335.2 | 52.5 | 225.8 | 29.5 | 0.39 |
| miR-4521 | 3.85 | 6.3 | 17.5 | 35.1 | 12.1 | 17.5 | 9.7 | 8.3 | 19.1 | 12.2 | 86.2 | 110.7 | 44.7 | 62.0 | 9.5 | 80.1 | 0.00 |
| miR-486-3p | 3.21 | 51.9 | 106.9 | 172.7 | 92.3 | 34.5 | 49.3 | 64.9 | 112.2 | 141.6 | 414.9 | 301.5 | 274.4 | 134.0 | 506.0 | 136.6 | 0.00 |
| miR-155 | 3.03 | 2267.3 | 2209.7 | 2711.0 | 2123.7 | 2827.9 | 2695.2 | 2055.6 | 2766.2 | 2636.6 | 3604.5 | 13339.1 | 11085.7 | 7526.8 | 5661.0 | 7490.1 | 0.00 |
| miR-1973 | 3.02 | 74.7 | 47.2 | 52.1 | 52.7 | 41.4 | 40.6 | 49.1 | 53.2 | 38.5 | 362.2 | 201.8 | 198.0 | 94.2 | 75.7 | 102.6 | 0.00 |
| miR-4524 | 2.83 | 2.1 | 1.7 | 1.7 | 2.2 | 2.3 | 2.6 | 2.3 | 2.5 | 1.6 | 1.9 | 11.1 | 7.8 | 7.3 | 4.8 | 7.1 | 0.00 |
| miR-1291 | 2.76 | 1.8 | 5.1 | 9.8 | 6.3 | 4.6 | 4.5 | 3.8 | 3.9 | 7.3 | 19.5 | 7.6 | 18.5 | 10.0 | 13.4 | 14.0 | 0.00 |
| miR-4304 | 2.68 | 2.1 | 2.6 | 2.2 | 1.3 | 2.5 | 1.8 | 1.5 | 2.3 | 1.7 | 10.0 | 4.5 | 3.8 | 2.8 | 18.2 | 2.4 | 0.00 |
| miR-550a | 2.67 | 1.6 | 6.8 | 4.0 | 8.8 | 5.5 | 6.1 | 6.6 | 8.6 | 7.0 | 23.2 | 22.0 | 13.5 | 10.4 | 16.4 | 9.3 | 0.00 |
| miR-550a-star | 2.44 | 3.4 | 6.1 | 4.9 | 8.3 | 7.2 | 8.9 | 6.9 | 12.5 | 7.5 | 14.8 | 29.3 | 17.5 | 11.1 | 23.5 | 11.1 | 0.00 |
| miR-595 | 2.41 | 8.3 | 8.7 | 2.7 | 4.4 | 2.8 | 5.6 | 4.9 | 5.3 | 2.7 | 8.4 | 10.9 | 10.0 | 7.9 | 27.8 | 9.3 | 0.00 |
| miR-331-5p | 2.41 | 6.3 | 8.4 | 9.1 | 12.4 | 11.2 | 10.1 | 9.2 | 8.3 | 11.0 | 41.4 | 20.9 | 13.9 | 17.9 | 37.2 | 16.8 | 0.00 |
| miR-21-star | 2.24 | 3.6 | 7.7 | 4.5 | 6.2 | 7.1 | 9.2 | 10.0 | 7.4 | 3.7 | 29.0 | 15.9 | 11.2 | 10.9 | 9.7 | 13.0 | 0.00 |
| miR-4793-3p | 2.23 | 7.5 | 7.7 | 45.4 | 12.0 | 12.4 | 8.4 | 11.2 | 10.2 | 18.4 | 49.2 | 19.3 | 24.4 | 12.4 | 48.0 | 32.0 | 0.64 |
| miR-34a-star | 2.22 | 2.2 | 1.9 | 2.8 | 2.7 | 2.0 | 2.0 | 2.8 | 2.2 | 3.1 | 4.7 | 1.9 | 18.7 | 3.1 | 17.3 | 2.4 | 1.05 |
| miR-574-3p | 2.21 | 71.6 | 87.4 | 103.5 | 89.9 | 67.0 | 88.9 | 110.1 | 117.5 | 82.0 | 155.1 | 336.0 | 210.3 | 158.0 | 238.7 | 145.6 | 0.00 |
| miR-486-5p | 2.19 | 148.9 | 295.1 | 313.3 | 228.7 | 138.5 | 144.9 | 207.7 | 272.7 | 276.2 | 941.2 | 434.0 | 425.9 | 274.5 | 817.3 | 278.0 | 0.00 |
| miR-4484 | 2.09 | 31.8 | 27.9 | 55.9 | 31.6 | 30.1 | 23.0 | 25.6 | 22.0 | 31.5 | 103.7 | 62.7 | 85.7 | 56.4 | 42.5 | 44.7 | 0.00 |
| miR-335 | 2.06 | 13.3 | 14.4 | 10.7 | 11.0 | 15.6 | 19.9 | 12.7 | 14.9 | 12.1 | 7.3 | 49.8 | 38.5 | 35.5 | 57.6 | 17.1 | 0.64 |
| miR-200a | 0.42 | 7.7 | 10.3 | 10.1 | 12.8 | 7.3 | 8.2 | 7.4 | 5.5 | 11.0 | 3.0 | 7.6 | 3.1 | 5.5 | 3.3 | 1.9 | 0.66 |

| | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| miR-181d | 0.24 | 42.0 | 67.7 | 43.2 | 60.4 | 54.6 | 44.0 | 46.0 | 44.8 | 44.8 | 5.2 | 12.0 | 25.3 | 24.6 | 2.1 | 33.4 | 0.66 |
| miR-363 | 0.22 | 290.0 | 340.2 | 378.6 | 483.0 | 341.1 | 326.9 | 286.1 | 342.9 | 351.7 | 149.8 | 96.7 | 122.0 | 248.4 | 10.1 | 39.4 | 0.66 |
| miR-451 | 0.19 | 65.2 | 270.3 | 40.9 | 95.4 | 106.3 | 115.1 | 118.9 | 106.5 | 69.9 | 7.8 | 9.6 | 46.2 | 50.6 | 8.1 | 28.5 | 0.00 |
| miR-181b | 0.15 | 301.6 | 623.1 | 602.7 | 971.2 | 640.4 | 614.3 | 559.4 | 577.1 | 582.6 | 99.9 | 15.4 | 358.9 | 216.3 | 7.9 | 510.3 | 4.91 |
| miR-181c | 0.13 | 42.4 | 76.9 | 160.2 | 228.6 | 148.5 | 131.1 | 117.7 | 113.9 | 118.9 | 13.5 | 2.1 | 48.2 | 40.2 | 2.7 | 72.7 | 0.66 |
| miR-181a | 0.11 | 1565.3 | 2632.8 | 3260.2 | 4689.1 | 2634.1 | 2494.3 | 2341.5 | 2016.6 | 2343.4 | 412.0 | 46.9 | 1106.1 | 733.6 | 18.2 | 1616.9 | 1.05 |
| miR-138 | 0.06 | 141.1 | 112.1 | 97.0 | 106.8 | 79.7 | 76.2 | 99.5 | 84.8 | 102.2 | 55.8 | 2.2 | 7.3 | 3.4 | 2.7 | 5.3 | 0.00 |

**Note**: C/N: ratio of miR expression in CLL cells/miR expression in normal B lymphocytes; N1-N9, Normal lymphocytes from 9 healthy Asian individuals; C1-C6, CLL cells from 6 Asian CLL patients.

**Supplementary Table 3. Differential microRNA expression in CLL cells and normal B lymphocytes from Western individuals**

| MicroRNA | Fold Change (CLL/Norm) | Western Samples | | | | | | | | | | | | | | | q-value (%) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | C1 | C2 | C3 | C4 | C5 | C6 | C7 | C8 | C9 | N1 | N2 | N3 | N4 | N5 | N6 | |
| miR-1295 | 31.78 | 246.6 | 109.8 | 34.4 | 160.8 | 99.0 | 114.9 | 34.5 | 5.1 | 84.1 | 1.9 | 2.0 | 1.2 | 1.7 | 4.0 | 2.7 | 0.00 |
| miR-451 | 28.27 | 636.0 | 43.9 | 57.0 | 88.9 | 7.3 | 57.7 | 1.8 | 582.9 | 109.7 | 1.5 | 2.0 | 2.1 | 2.5 | 3.4 | 1.4 | 0.00 |
| miR-4793-3p | 27.73 | 6.1 | 456.1 | 647.4 | 215.5 | 38.6 | 234.7 | 807.9 | 320.7 | 141.2 | 9.1 | 2.6 | 2.3 | 12.3 | 3.9 | 20.7 | 0.00 |
| miR-4440 | 17.53 | 4.9 | 326.6 | 539.0 | 145.8 | 38.2 | 208.5 | 739.1 | 315.4 | 150.9 | 9.3 | 2.3 | 9.5 | 19.7 | 5.2 | 18.5 | 0.00 |
| miR-210 | 6.27 | 137.4 | 363.5 | 184.6 | 186.7 | 46.1 | 551.2 | 63.1 | 81.2 | 319.8 | 22.4 | 40.4 | 38.6 | 18.3 | 26.4 | 16.7 | 0.00 |
| miR-185-star | 6.25 | 3.7 | 26.9 | 24.1 | 36.1 | 5.2 | 20.5 | 81.3 | 60.7 | 18.5 | 3.8 | 2.3 | 1.9 | 6.7 | 3.1 | 4.3 | 0.00 |
| miR-148a | 4.82 | 54.2 | 56.1 | 12.8 | 46.1 | 70.4 | 57.1 | 22.5 | 7.6 | 8.0 | 2.8 | 2.7 | 9.7 | 25.3 | 2.4 | 8.3 | 0.82 |
| miR-3197 | 4.65 | 121.7 | 24.0 | 28.5 | 284.0 | 127.0 | 197.2 | 44.8 | 10.3 | 164.1 | 21.1 | 12.6 | 13.8 | 28.5 | 11.9 | 10.0 | 0.82 |
| miR-4524-star | 4.50 | 5.0 | 2.8 | 4.6 | 18.2 | 8.9 | 14.6 | 2.0 | 4.8 | 5.7 | 1.3 | 1.5 | 1.2 | 1.4 | 1.2 | 1.3 | 0.00 |
| miR-3907 | 4.29 | 4.7 | 28.2 | 32.7 | 17.2 | 4.7 | 27.8 | 54.9 | 37.6 | 9.8 | 2.5 | 2.5 | 5.1 | 3.7 | 5.5 | 8.7 | 0.00 |
| miR-3148 | 4.27 | 1.7 | 16.9 | 13.0 | 7.5 | 3.5 | 20.5 | 50.6 | 17.9 | 5.3 | 3.3 | 2.2 | 2.4 | 2.0 | 2.3 | 2.0 | 0.82 |
| miR-4717-3p | 4.17 | 5.4 | 30.8 | 31.2 | 17.2 | 6.8 | 31.6 | 107.7 | 44.7 | 13.0 | 6.0 | 6.3 | 5.6 | 5.4 | 3.4 | 5.6 | 0.00 |
| miR-486-5p | 3.53 | 1328.7 | 444.7 | 1014.6 | 508.7 | 263.9 | 428.4 | 264.4 | 2509.7 | 1165.6 | 238.5 | 103.8 | 279.0 | 214.2 | 458.6 | 67.7 | 0.82 |
| miR-4496 | 3.49 | 20.7 | 22.6 | 29.2 | 38.8 | 21.6 | 18.5 | 94.2 | 68.0 | 40.6 | 6.4 | 11.2 | 12.7 | 3.6 | 29.8 | 8.1 | 0.00 |
| miR-4462 | 3.47 | 14.8 | 54.2 | 75.1 | 51.2 | 17.7 | 60.0 | 189.0 | 64.0 | 32.9 | 12.3 | 7.2 | 12.2 | 16.0 | 21.0 | 18.6 | 0.00 |
| miR-4778-5p | 3.15 | 20.9 | 34.7 | 19.7 | 17.2 | 19.1 | 28.3 | 30.4 | 12.4 | 28.5 | 14.7 | 13.8 | 6.1 | 4.6 | 4.3 | 5.3 | 0.00 |
| miR-3135b | 3.07 | 493.8 | 982.9 | 2759.9 | 3461.6 | 3888.1 | 1417.2 | 3238.5 | 635.8 | 1878.5 | 954.8 | 607.2 | 591.5 | 664.9 | 434.1 | 258.4 | 0.82 |
| miR-4486 | 2.93 | 45.4 | 158.4 | 193.8 | 112.4 | 137.9 | 168.1 | 306.4 | 163.9 | 147.8 | 119.4 | 63.8 | 64.1 | 35.1 | 26.8 | 31.0 | 0.00 |
| miR-155 | 2.76 | 6030.1 | 7503.2 | 10732.7 | 7729.2 | 10618.7 | 8300.3 | 6429.5 | 4340.3 | 7667.9 | 2982.4 | 2919.8 | 2722.0 | 2403.4 | 2279.7 | 2949.3 | 0.00 |
| miR-3178 | 2.48 | 349.7 | 449.1 | 801.1 | 552.8 | 383.1 | 706.9 | 977.8 | 656.4 | 369.0 | 196.7 | 314.5 | 187.3 | 169.6 | 182.4 | 328.1 | 0.00 |
| miR-4507 | 2.47 | 1001.4 | 949.8 | 1859.8 | 1498.2 | 943.8 | 1084.2 | 2542.6 | 580.8 | 1307.3 | 1005.3 | 439.9 | 485.1 | 279.0 | 330.8 | 673.0 | 0.00 |
| miR-4745-5p | 2.47 | 280.8 | 370.0 | 608.3 | 1274.2 | 738.7 | 721.8 | 514.5 | 291.7 | 457.3 | 264.5 | 327.4 | 246.8 | 116.2 | 195.4 | 188.3 | 0.00 |
| miR-4530 | 2.43 | 955.0 | 1084.4 | 2360.1 | 3292.2 | 1459.6 | 1640.1 | 2082.4 | 824.6 | 1099.4 | 1030.3 | 644.6 | 742.3 | 387.7 | 400.6 | 695.4 | 0.00 |
| miR-4322 | 2.39 | 16.0 | 14.7 | 7.5 | 41.0 | 20.7 | 17.3 | 10.3 | 7.0 | 15.9 | 4.6 | 11.5 | 10.0 | 4.2 | 4.4 | 5.2 | 1.21 |

| | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| miR-1915 | 2.38 | 1389.4 | 1537.2 | 2586.1 | 5785.5 | 3117.3 | 2821.5 | 2351.5 | 1227.9 | 2456.2 | 1501.0 | 1223.5 | 1119.0 | 518.4 | 850.7 | 972.1 | 0.00 |
| miR-4485 | 2.34 | 2450.4 | 4780.3 | 9210.7 | 3264.0 | 3585.6 | 1601.6 | 4016.6 | 6149.4 | 3047.4 | 2193.0 | 2234.4 | 1851.0 | 1326.5 | 922.9 | 1561.5 | 0.82 |
| miR-3187-3p | 2.31 | 9.9 | 4.5 | 16.0 | 33.3 | 29.5 | 11.7 | 26.3 | 14.4 | 8.5 | 6.8 | 10.7 | 10.0 | 4.3 | 4.0 | 4.6 | 2.91 |
| miR-2861 | 2.25 | 973.1 | 1347.1 | 2633.1 | 4376.0 | 2384.3 | 2146.9 | 2087.0 | 1025.0 | 1864.9 | 1264.2 | 1119.3 | 1079.9 | 466.7 | 644.1 | 757.0 | 0.82 |
| miR-483-5p | 2.25 | 51.0 | 57.7 | 42.4 | 47.8 | 55.8 | 57.5 | 88.2 | 27.9 | 50.3 | 59.9 | 35.6 | 16.0 | 22.2 | 10.4 | 17.3 | 0.82 |
| miR-4497 | 2.22 | 2346.2 | 2502.0 | 3705.7 | 4377.4 | 3092.0 | 3042.4 | 3536.9 | 2595.9 | 2572.7 | 2284.2 | 1395.2 | 1227.9 | 856.1 | 1180.5 | 1608.4 | 0.00 |
| miR-1587 | 2.19 | 427.7 | 334.7 | 701.3 | 677.2 | 448.6 | 374.7 | 1285.7 | 156.0 | 666.0 | 536.5 | 190.7 | 185.2 | 124.3 | 168.3 | 300.1 | 3.82 |
| miR-4763-3p | 2.15 | 737.4 | 975.2 | 1647.6 | 1990.2 | 1207.6 | 1200.2 | 1833.4 | 558.1 | 1199.3 | 1056.2 | 614.7 | 612.9 | 283.9 | 440.9 | 530.8 | 0.82 |
| miR-3646 | 2.09 | 6.5 | 10.4 | 24.7 | 11.9 | 8.5 | 11.7 | 15.2 | 6.3 | 13.6 | 4.4 | 5.5 | 6.3 | 4.7 | 4.0 | 7.7 | 0.00 |
| miR-4505 | 2.08 | 562.1 | 390.7 | 1057.1 | 1370.9 | 705.5 | 691.4 | 1436.5 | 228.8 | 783.5 | 530.5 | 218.4 | 291.2 | 271.6 | 290.4 | 537.4 | 3.38 |
| miR-3127-5p | 2.07 | 7.5 | 6.9 | 5.7 | 5.3 | 11.7 | 8.9 | 6.0 | 3.4 | 13.3 | 8.1 | 5.6 | 2.1 | 2.9 | 2.8 | 2.1 | 2.64 |
| miR-1306 | 2.00 | 7.2 | 8.1 | 5.0 | 4.3 | 5.5 | 4.0 | 14.5 | 8.4 | 5.8 | 4.3 | 4.2 | 3.4 | 2.6 | 3.4 | 2.1 | 0.82 |
| miR-422a | 0.48 | 48.3 | 21.7 | 31.6 | 24.5 | 29.8 | 12.3 | 33.2 | 20.4 | 50.9 | 57.7 | 72.9 | 28.3 | 107.4 | 39.2 | 77.9 | 4.26 |
| miR-4288 | 0.48 | 8.9 | 4.8 | 2.0 | 7.6 | 3.9 | 3.7 | 5.2 | 5.1 | 3.4 | 6.1 | 10.0 | 7.5 | 12.1 | 11.1 | 12.2 | 1.56 |
| miR-1275 | 0.48 | 426.7 | 345.7 | 232.0 | 225.2 | 400.7 | 243.8 | 211.3 | 227.6 | 184.5 | 960.0 | 869.5 | 653.2 | 241.7 | 585.8 | 400.1 | 1.56 |
| miR-320e | 0.47 | 109.1 | 59.3 | 95.6 | 119.6 | 144.4 | 58.1 | 117.4 | 62.7 | 311.6 | 304.3 | 208.4 | 181.1 | 234.0 | 236.9 | 189.3 | 2.17 |
| miR-629 | 0.45 | 48.4 | 99.3 | 18.1 | 35.2 | 47.1 | 42.0 | 40.6 | 31.2 | 80.9 | 90.6 | 149.7 | 83.0 | 88.8 | 98.6 | 93.4 | 1.09 |
| miR-221 | 0.43 | 1139.9 | 1088.8 | 648.5 | 1031.7 | 1068.8 | 1035.1 | 1406.0 | 989.3 | 899.8 | 2146.7 | 2233.9 | 2208.8 | 2591.8 | 2499.9 | 2618.9 | 0.00 |
| miR-181a-star | 0.42 | 1.8 | 2.8 | 3.0 | 8.8 | 1.6 | 2.5 | 2.8 | 2.8 | 2.4 | 2.4 | 6.3 | 10.6 | 11.5 | 5.5 | 8.4 | 3.07 |
| miR-652 | 0.41 | 176.6 | 108.1 | 103.7 | 84.0 | 122.3 | 113.9 | 120.0 | 112.9 | 108.8 | 235.7 | 289.0 | 241.2 | 371.7 | 279.7 | 265.2 | 0.00 |
| miR-222 | 0.39 | 1257.1 | 1553.1 | 1404.3 | 1882.3 | 1652.3 | 1397.4 | 1928.3 | 1713.7 | 1163.9 | 3495.5 | 4258.7 | 4280.3 | 2853.3 | 4714.4 | 3998.0 | 0.00 |
| miR-4521 | 0.38 | 51.9 | 38.3 | 59.8 | 53.3 | 72.4 | 20.2 | 18.5 | 57.4 | 18.3 | 147.1 | 162.6 | 64.0 | 42.6 | 143.7 | 111.2 | 2.17 |
| miR-484 | 0.38 | 27.1 | 16.0 | 9.1 | 12.5 | 12.3 | 19.5 | 16.7 | 32.2 | 40.0 | 30.2 | 53.8 | 51.6 | 80.5 | 52.0 | 39.3 | 0.00 |
| miR-3195 | 0.36 | 31.4 | 25.8 | 48.9 | 36.8 | 22.0 | 20.4 | 61.5 | 47.7 | 16.7 | 131.7 | 167.3 | 66.1 | 44.3 | 105.6 | 65.3 | 0.00 |
| miR-152 | 0.36 | 12.7 | 23.4 | 2.9 | 3.4 | 8.6 | 9.0 | 6.5 | 3.9 | 7.7 | 20.9 | 10.8 | 13.5 | 38.5 | 22.9 | 22.1 | 2.17 |
| miR-4443 | 0.35 | 192.8 | 199.1 | 183.3 | 258.3 | 222.0 | 226.1 | 323.9 | 496.9 | 391.6 | 1207.5 | 571.6 | 1062.5 | 778.3 | 757.1 | 438.7 | 0.00 |
| miR-378-star | 0.34 | 5.8 | 6.0 | 3.5 | 8.6 | 3.4 | 3.8 | 7.4 | 8.7 | 26.1 | 11.4 | 27.8 | 16.9 | 27.9 | 13.9 | 24.1 | 1.09 |
| miR-1973 | 0.30 | 89.7 | 56.5 | 74.7 | 53.1 | 45.7 | 64.3 | 165.4 | 73.0 | 51.1 | 303.6 | 200.7 | 281.4 | 227.0 | 189.3 | 203.0 | 0.00 |

| | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| miR-146a | 0.29 | 225.8 | 354.1 | 288.1 | 126.9 | 205.7 | 493.2 | 541.7 | 1394.3 | 521.0 | 787.8 | 1219.3 | 1493.3 | 1912.8 | 990.0 | 1495.0 | 0.00 |
| miR-720 | 0.25 | 332.6 | 184.5 | 69.2 | 84.7 | 107.4 | 95.5 | 681.9 | 223.8 | 38.5 | 932.9 | 679.5 | 367.5 | 455.6 | 358.2 | 785.0 | 1.09 |
| miR-1260 | 0.25 | 6.7 | 2.7 | 3.1 | 2.1 | 3.5 | 5.0 | 7.0 | 2.6 | 4.3 | 5.0 | 26.7 | 11.7 | 7.3 | 18.6 | 54.2 | 0.00 |
| miR-1280 | 0.19 | 125.5 | 76.4 | 46.9 | 49.8 | 45.3 | 82.8 | 131.4 | 150.4 | 57.2 | 616.7 | 392.1 | 265.0 | 306.5 | 325.9 | 731.0 | 0.00 |
| miR-1260b | 0.16 | 97.1 | 80.9 | 60.3 | 20.5 | 55.2 | 61.3 | 157.1 | 163.6 | 63.2 | 814.1 | 361.2 | 281.2 | 327.5 | 416.1 | 691.6 | 0.00 |
| miR-363 | 0.16 | 29.4 | 15.3 | 5.2 | 26.8 | 38.4 | 10.4 | 18.6 | 55.5 | 24.4 | 73.3 | 128.8 | 93.8 | 191.6 | 158.1 | 175.8 | 0.00 |
| miR-23a-star | 0.15 | 17.2 | 10.3 | 21.4 | 23.3 | 26.2 | 18.2 | 21.9 | 9.8 | 22.7 | 486.6 | 157.3 | 147.4 | 50.7 | 57.2 | 108.0 | 0.00 |
| miR-4286 | 0.14 | 26.0 | 4.8 | 5.6 | 12.4 | 6.5 | 4.3 | 7.8 | 27.0 | 3.8 | 94.0 | 77.0 | 28.1 | 80.6 | 29.1 | 83.5 | 0.00 |
| miR-151b | 0.13 | 379.0 | 22.5 | 2.8 | 164.7 | 24.3 | 69.1 | 8.4 | 453.6 | 21.5 | 199.2 | 256.1 | 347.7 | 310.7 | 559.6 | 364.8 | 4.33 |
| miR-3150b-3p | 0.12 | 2.8 | 3.0 | 2.4 | 3.2 | 3.2 | 1.6 | 2.6 | 2.6 | 2.5 | 25.4 | 24.0 | 30.5 | 10.1 | 22.6 | 20.0 | 0.00 |
| miR-193b | 0.11 | 2.9 | 7.6 | 7.6 | 1.4 | 2.3 | 6.3 | 3.2 | 9.6 | 14.5 | 62.6 | 47.8 | 67.2 | 35.1 | 30.1 | 31.1 | 0.00 |
| miR-181c | 0.10 | 3.9 | 3.0 | 2.5 | 2.1 | 3.5 | 3.0 | 20.0 | 3.0 | 4.6 | 17.7 | 63.5 | 40.1 | 28.9 | 93.5 | 28.1 | 0.00 |
| miR-181a | 0.08 | 162.7 | 246.4 | 287.8 | 41.9 | 75.2 | 279.5 | 1096.6 | 171.2 | 148.8 | 2730.5 | 2250.1 | 2889.1 | 2541.9 | 3323.3 | 1129.5 | 0.00 |
| miR-151-3p | 0.07 | 198.5 | 3.6 | 1.6 | 66.3 | 4.0 | 16.9 | 2.3 | 169.2 | 7.9 | 96.0 | 138.1 | 230.7 | 166.8 | 318.1 | 224.4 | 1.56 |
| miR-27a-star | 0.07 | 4.4 | 6.7 | 4.2 | 6.9 | 15.3 | 2.5 | 2.9 | 4.5 | 14.5 | 96.3 | 115.0 | 65.6 | 90.1 | 34.8 | 134.6 | 0.00 |
| miR-181b | 0.06 | 28.5 | 43.2 | 77.4 | 4.9 | 12.6 | 46.2 | 168.9 | 31.1 | 24.1 | 385.7 | 439.4 | 818.9 | 552.2 | 690.6 | 357.1 | 0.00 |
| miR-138 | 0.06 | 2.1 | 2.0 | 1.4 | 1.8 | 2.5 | 1.7 | 1.2 | 1.9 | 16.1 | 74.3 | 107.9 | 26.5 | 6.3 | 19.9 | 133.3 | 0.00 |
| miR-181d | 0.43 | 4.04 | 2.20 | 3.16 | 1.42 | 2.96 | 3.58 | 2.77 | 4.80 | 4.22 | 3.67 | 3.29 | 21.32 | 16.72 | 4.19 | 7.52 | 3.01 |

**Note:** C/N: ratio of miR expression in CLL cells/miR expression in normal B lymphocytes; C1-C9, CLL cells from 9 Western CLL patients; N1-N6, Normal lymphocytes from 6 healthy Western individuals.

**Supplementary Table 4. Changes in genes expression in CLL cells and normal B lymphocytes from GSE66117 dataset.**

| Gene name | Fold Change (CLL/Normal) | *P* Value |
|---|---|---|
| *TGR5* | 0.437 | 0.025 |
| *ASB7* | 1.033 | 0.771 |
| *CTDSPL2* | 1.275 | 0.033 |
| *FBXO32* | 1.595 | 0.021 |
| *JOSD1* | 0.646 | 0.054 |
| *MED1* | 0.973 | 0.778 |

**Note:** Gene expression of more than 2-fold change (FC > 2.0 or < 0.5) with a *P* value less than 0.05 was considered statistically significant. Only TGR5 met this criterion; CLL, n=45; normal, n=5.

**Supplementary Table 5. List of antibodies used for Western blot analysis.**

| Antibody Name | Species | Catalog# | Manufacturer |
|---|---|---|---|
| Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP® mAb | Rabbit | #4370 | Cell Signaling Technology Inc. |
| p44/42 MAPK (Erk1/2) (137F5) mAb | Rabbit | #4695 | Cell Signaling Technology Inc. |
| G-protein coupled bile acid receptor 1 | Rabbit | YT4636 | Immunoway |
| Bcl-2 (D17C4) mAb | Rabbit | #3498 | Cell Signaling Technology Inc. |
| Bcl-xL Antibody | Rabbit | #2762 | Cell Signaling Technology Inc. |
| Stat3 (D3Z2G) mAb | Rabbit | #12640 | Cell Signaling Technology Inc. |
| Phospho-Stat3 (Tyr705) (D3A7) XP® mAb | Rabbit | #9145 | Cell Signaling Technology Inc. |
| MEK1/2 Antibody | Rabbit | #9122 | Cell Signaling Technology Inc. |
| Phospho-MEK1/2 (Ser217/221) (41G9) Rabbit mAb | Rabbit | #9154 | Cell Signaling Technology Inc. |
| c-Jun (60A8) mAb | Rabbit | #9165 | Cell Signaling Technology Inc. |
| Phospho-c-Jun-S63 pAb | Rabbit | AP0048 | ABclonal |
| Vinculin (E1E9V) XP® mAb | Rabbit | #13901 | Cell Signaling Technology Inc. |
| GAPDH (14C10) mAb | Rabbit | #2118 | Cell Signaling Technology Inc. |