Front. Biol. 2016, 11(1): 53–58
DOI 10.1007/s11515-016-1387-1

RESEARCH ARTICLE

Evaluation of microRNA-146a expression in acute lymphoblastic leukemia

Farzaneh Tavakoli[1], Kaveh Jaseb[1], Mohammad Ali Jalali Far[1], Masoud Soleimani[2], Elahe Khodadi[1], Najmaldin Saki (✉)[1]

[1] *Health Research Institute, Research Center of Thalassemia & Hemoglobinopathy, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran*
[2] *Department of hematology, School of Medical Sciences, Tarbiat Modares University, Tehran, Iran*

Abstract **MicroRNAs (miRNAs) play an essential role in the development and progression of acute lymphoblastic leukemia (ALL), and could serve as disease biomarkers and therapeutic targets. The function of miR-146a in lymphoid differentiation has been here with discussed. However, the role of this miRNA in the outcome of ALL is not well understood. Peripheral blood of 48 patients with ALL and 20 age- and sex-matched healthy control subjects was used to accurately evaluate the expression of miR-146a by stem-loop Real time PCR. No statistically significant difference was found between patients and controls in total miR-146a expression. The expression of miR-146a was high (18.75%), low (27.08%) and not different (54.17%) in ALL patients. Our analysis indicated no association between the expression of miR-146a and any prognostic factors such as WBC/PLT counts, Hb, fusion genes (P190 and some translocations) with ALL type. This study revealed that miR-146a cannot be an independent factor for predicting the outcome of ALL patients. We suggest a multi-parameter analysis including miRNAs, transcription factors and critical genes to achieve a precise clinical panel for prognostic values.**

Keywords miR-146a, acute lymphoblastic leukemia, prognostic marker

Introduction

Acute lymphoblastic leukemia (ALL) is the most common malignancy in children and accounts for nearly 25% of malignancies diagnosed among children under 15 years of age (Mei et al., 2014). In this leukemia, progenitor lymphoid cells proliferate and replace the normal hematopoietic cells of the bone marrow. This causes reduced production of normal blood cells (Hagag et al., 2014). Extramedullary manifestations can also be observed in central nervous system (CNS), lymph nodes, gonads, spleen, and liver (Portell et al., 2013). Studies showed that crucial sequential events (specific chromosomal translocations or fusion genes) can be the first hits to initiate ALL and further genetic or epigenetic events (gene deletions or mutations) are the second hits involved in the outbreak of ALL (Du et al., 2013). Recently, many studies suggest that microRNAs can play a substantial role in leukemogenesis (Schotte et al., 2010; Bottoni and Calin, 2013; Yan et al., 2013; Yin et al., 2014).

MiRNAs are 20-22 nucleotide non-coding RNA molecules playing a vital role in control of critical cellular processes such as proliferation and differentiation through post translational regulation (Hauptman and Glavac, 2013). Degradation of target mRNA occurs when miRNA and its target mRNA are complementary to each other exactly (perfect match) or nearly exactly (Faraoni et al., 2009). Some miRNAs are stable in serum or plasma and show unique expression patterns; therefore, they can be used as biomarkers for various diseases (Xiong et al., 2014). Studies have indicated the likelihood of distinguishing between malignant B cells from normal B cells as well as activated B cells from inactivated ones by miRNA expression profiles (Li and Wang, 2013). There have been increasing studies on the relationship between adult cancers and miRNAs in recent years but there are still a relatively low number of studies regarding ALL (Duyu et al., 2014). Particular miRNA signatures such as miR-124-1, let-7a-3, miR-181, miR-29B and their role in pathogenesis, diagnosis

Received December 16, 2015; accepted January 26, 2016
Correspondence: Najmaldin Saki
E-mail: najmaldinsaki@gmail.com

and prognosis of myeloid and lymphoid leukemias have been discovered (Yin et al., 2014).

MiRNA-146a was first noticed for its activity in innate immune system. Then, its role in acquired immunity was described. Loss of miRNA-146a in mice causes hematological disorders (So et al., 2013). The pattern of miRNA-146a alternation in patients with leukemia has been analyzed and the results were noticeable. Also, the association between this miRNA and some genetic abnormalities in malignancies has been demonstrated (Table 1). However, evaluation of this miRNA in ALL seems to be insufficient and demands more investigations. In view of this evaluation, the prognostic parameters (including age, gender, WBC/Plt counts, Hb, lineages and cytogenetic abnormalities) have been considered in this study. So, we try to focus on this issue and find out the relationship between miR-146a and clinical outcome of ALL patients.

Materials and methods

Patients and samples

ALL was diagnosed via BM aspirate displaying at least 30% blast cells based on FAB classification. Following morphologic, cytogenetic and clinical examinations on 48 ALL patients, the subjects were enrolled in this study, including 31 males (64.6%) and 17 females (35.4%) (1–26 years old; median age: 8.05 years). Forty- five cases (93.75%) were B-lineages and 3 cases (6.25%) were T-lineages ALL. Twenty age-and sex-matched healthy control subjects with no morphologic, cytogenetic and clinical disorders were also enrolled in this study. Table 2 shows patients' information. All human peripheral blood samples were obtained with written informed consent from the subjects. This study was approved by the local ethics committee of Ahvaz Jundishapur University of Medical Sciences (AJUMS.REC.1393.310).

Cell culture

Jurkat (suspension, lymphoblast-like, obtained from acute lymphoblastic leukemia; ALL) and U937 (suspension, monocyte-like, derived from histiocytic lymphoma lymphocyte and myeloid cells) cell lines were purchased from Pasture Institute of Iran and were maintained in RPMI-1640 medium (Gibco, Carlsbad, CA, USA) containing 2 mmol/L glutamine, 25mmol/L HEPES, 1.5g/L sodium bicarbonate, 10% fetal calf serum (FCS), 50 U/mL penicillin and 50 g/mL streptomycin at 37°C in 5% CO_2.

MiRNAs extraction, cDNA synthesis, and real-time polymerase chain reaction

Total miRNA was extracted from blood samples using RNX Plus™ solution (Cinnagen, Iran) according to manufacturer's instructions. The qualification of extracted miRNAs was performed by measuring the absorbance at 260 nm. cDNA synthesis was performed as follows: 1.5 µL of specific primer, 3 µL RNA and 15.5 µL distilled water were mixed. Then, the mixture was placed in ABI step one plus PCR instrument for 5 min in 95°C and 10 min in 70°C. After completing this step, 3µL RT buffer, 3µL dNTP, 1µL RT enzyme and 3µL distilled water was added to this mixture for 15 min at 25°C, 15 min at 37°C, 60 min in 42°C and 10 min at 75°C in the instrument, respectively. MiRNA was quantified by realtime polymerase chain reaction (PCR) with 4 µL Fermentas SYBR Green Mastermix (Fermentas Life Sciences, St Leon-Rot, Germany) in a Rotor-Gene 6000 system (Corbett, Concorde, NSW, Australia) according to manufacturer's instructions. TaqMan® MicroRNA Reverse Transcription Kit (Applied Biosystems, Grand Island, NY, USA) was used for reverse transcription with 2 µL target specific stem loop primers provided in the TaqMan® miRNA assays. Then, 2 µL of template and 12 µL of distilled water were added. The reactions were performed as follows: initial polymerase activation at 95°C for 15 min. For miRNA quantification, 40 amplification cycles at 95°C for 10 s, 58°C for 20 s, and 72°C for 20 s with fluorescence detection were performed. The following primers were used for real time PCR: mir-146a RT: GTCGTATGCAGAGCAGGGTCCGAGGTATTCGCA CTGCATACGACAACCCAT, miR-146a Forward: TCCGT GAGAACTGAATTCC; miR-146a reverse: GAGCAGGGT CCGAGGT. For miRNA, the data were normalized using the small nucleolar RNA, C/D box (snord) endogenous

Table 1 Association between the expression of miR-146a and genetic abnormalities in leukemia

Malignancies	Genetic abnormalities	Target genes	Prognosis	References
AML	t(15;17)	*PML-RARA*	Favorable	Daschkey et al., 2013
AML	t(8;21)	*RUNX1-RUNX1T1*	Favorable	Daschkey et al., 2013
CML	t(9;22)	*BCR–ABL*	—	Ferreira et al., 2014
ALL	t(1;19)	*TEL-AML1*	—	Schotte et al., 2011
CLL	+ 12	*ITGA4, ITGB2*	Unfavorable	Visone et al., 2009
MDS	−5q	*CDC25C,PP2a*	Different	Ebert 2010; Starczynowski et al., 2010; Gaballa and Besa, 2014

Abbreviation: AML: acute myeloid leukemia; CML: chronic myeloid leukemia; ALL: acute lymphoblastic leukemia; CLL: chronic lymphoblastic leukemia; MDS: myelodysplastic syndrome; *PML-RARA*: Promyelocytic leukemia-retinoic acid receptor α; *RUNX1-RUNX1T1*: Runt-related transcription factor 1; *BCR–ABL*: breakpoint cluster region- Abelson murine leukemia viral oncogene homolog; *TEL-AML1*: Telomere length regulation protein- 1-acute myeloid leukemia 1; *ITGA4*: integrin subunit α 4; *ITGB2*: integrin subunit β 2; *CDC25C*: cell division cycle 25C; *PP2a*: Protein phosphatase 2.

control. Primers for snord 47 RT: GTCGTATGCAGAGCAG-GGTCCGAGGTATTCGCACTGCATACGACCACCTC, snord 47 F: ATCACTGTAAAACCGTTCA, were prepared based on the sequence derived from the study of Naderi and colleagues (Naderi et al., 2015). After cDNA synthesis, miR-146a RNA was used to provide a standard curve. The first dilution of the standard included 1500000 copies/mL of miR-146a cDNA. 5-fold serial dilution was applied to provide the standard curve.

Statistical analysis

Data were analyzed using Statistical Package for the Social Sciences (SPSS version 13.0). Each experiment was repeated at least three times. REST software (2009, QIAGEN, Valencia, USA) was used to analyze relative gene expression data of real-time PCR. $p \leqslant 0.01$ was considered to be statistically significant.

Results

MiR-146a expression

No statistically significant difference was found between miR-146a expression in patients and controls. Total expression of this miRNA was not different in our cell lines compared to controls. Mir-146a expression in U937cell line was 2.2 times higher than Jurkat cell line (Fig. 1).

MiR-146a expression, clinico-hematological and molecular studies

Among 48 patients enrolled in this study, miR-146a expression was high (18.75%), low (27.08%), and not different (54.17%). We compared miR-146a expression and some prognostic characteristics of patients, including age, gender, WBC/Plt counts, Hb, lineages and cytogenetic abnormalities. There was no significant relationship in this regard (Table 2) (Table 3) ($p < 0.05$). Overall, these results indicate that miR-146a is not a suitable independent prognostic factor in ALL patients.

In our study, among 48 patients, 3 patients were BCR-ABL positive, 3 patients were T-ALL, 2 patients were 11q23 positive and one patient showed t(1;14) as a rare chromosomal abnormality. Our finding showed no significant relationship in the expression of miR-146a in these patients. We also investigated other prognostic factors, including WBC and PLT counts, Hb and organomegaly and found no association

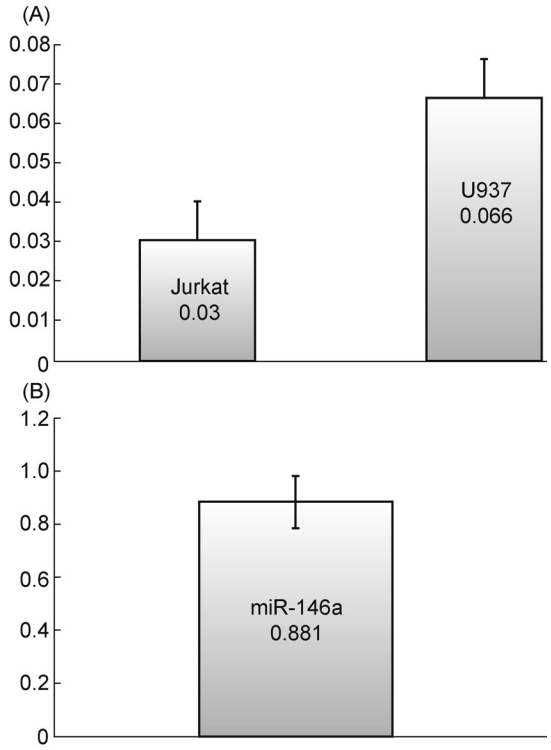

Figure 1 (A) miR-146a expression in ALL patients, (B) miR-146a expression in Jurkat and U937 cell lines.matrix metallopeptidase 16, MT3: metallothionein 3.

between the level of miR-146a and these parameters for distinguishing subtypes or predicting the clinical outcome. Following up our patients for 14 months, we had 2 deceased patients, one of whom was T-ALL with no difference in the expression of miR-146a compared to control group. The other had a high expression level of miR-146a. We observed more invasive clinical features in patients with low expression, which was not statistically significant.

Discussion

ALL, the most common malignancy in children, has achieved remarkable treatment improvements by virtue of recent molecular biomarkers (Friedmann and Weinstein, 2000). Experimental works have shed light on the diagnostic and prognostic role of miRNAs in hematologic disorders (Han et al., 2011; Li et al., 2011; de Oliveira et al., 2012). MiR-146a is implicated in the pathology of leukemia in an NF-κB-dependent manner as well as targeting CXCR4 and smad4

Table 2 Correlation between miR-146a expression and prognostic characteristics of ALL patients

Log expression		WBC	HB	PLT	RBC	Age	Treatment duration
	Correlation Coefficient	0.14	−0.14	0.00	−0.11	−0.16	−0.20
	p-value	0.35	0.33	1.0	0.46	0.92	0.18

Table 3 MiR-146a expression levels based on sex, type and BCR

		Expression	*p* value
Sex	Male (n = 31)	0.239 (0.001 – 1763.77)	0.56
	Female (n = 17)	1.783 (0.001 – 59658.68)	
Type	B-ALL (n = 45)	1.017 (0.001 –59658.68)	0.59
	T-ALL (n = 3)	0.517 (0.011 – 1.453)	
BCR	Negative (n = 45)	1.017 (0.001 –59658.68)	0.30
	Positive (n = 3)	0.517 (0.011 – 2.769)	

(Fig. 2) (Ferreira et al., 2014; Jurado-Camino et al., 201). Demonstrating the impact of miR-146a on lymphocyte differentiation, we selected this immune miRNA to figure out its role in the outcome of leukemia patients.

Mavrakis et al. (2011) showed overexpression of miR-146a in T-ALL patients, which was not consistent with our observation, as we had 3 cases of T-ALL, one of which had low miR-146 aexpression and the other two were not different with controls. Muhterem Duyu et al. conducted a follow-up study and their results demonstrated a high level of miR-146a at diagnosis, which was reduced after 6 months of treatment (Duyu et al., 2014). This result can indicate a seemingly oncogenic role of this miRNA. Consistent with this study, a recent report by Zhang et al. (2009) indicated the over-expression of miR-146a in 49 children with ALL. Also, another analysis stated that a high level of miR-146a can predict unfavorable outcome in adult ALL patients (Wang et al., 2010). However, non-clinical-trial studies showed opposite results. Data showed a tumor suppressive role of miR-146a through targeting early growth response 1 (EGR-1) protein (Contreras et al., 2015). In line with this, P Rosato et al. (2012) suggested that EBNA2 induces B cell transformation by reducing the expression of miR-146a, supporting the anti-leukemic role of this miRNA.

Fallah and colleagues showed that miR-146a induces upregulation of Ikaros, CD2, CD4, CD25 in CD133 + cells, causing differentiation into T lymphoid lineage (Fallah et al., 2013). In contrast, we concluded that this miRNA has no differentiation effect upon T cell lymphoblasts in our previous study by observing upregulation of PU-1 and downregulation of Notch1, ETS2, Ikaros, SOS1, CD3, CD4, CD8, CD25, and TCRα by MiR-146a (Saki et al., 2014). It seems that this miRNA has a basic role during differentiation of lymphocytes and its function depends on the expression and repression of other genes, transcription factors and miRNAs. Taken together, our findings reveal that miR-146a cannot be an independent prognostic factor for ALL patients. Some studies have shown the correlation between miR-146a and miR-155 and increased expression of them in inflammatory disease but not with other miRNAs (Béres et al., 2016). The expression of MiR-146a and miR-150 genes is also regulated by p53 and NFκB p65/RelA in human cervical carcinoma (Ghose and Bhattacharyya, 2015). In addition, correlation between miR-155 and mir-150 in neoplastic B cell development has been approved in some studies (Musilova and Mraz, 2015). So, this can suggest a correlation between miR-146a, 155 and 150 in malignancies. In conclusion, simultaneous evaluation of miR-146a, miR-155 and miR-150 for future studies in this field can be suggested for better prognosis.

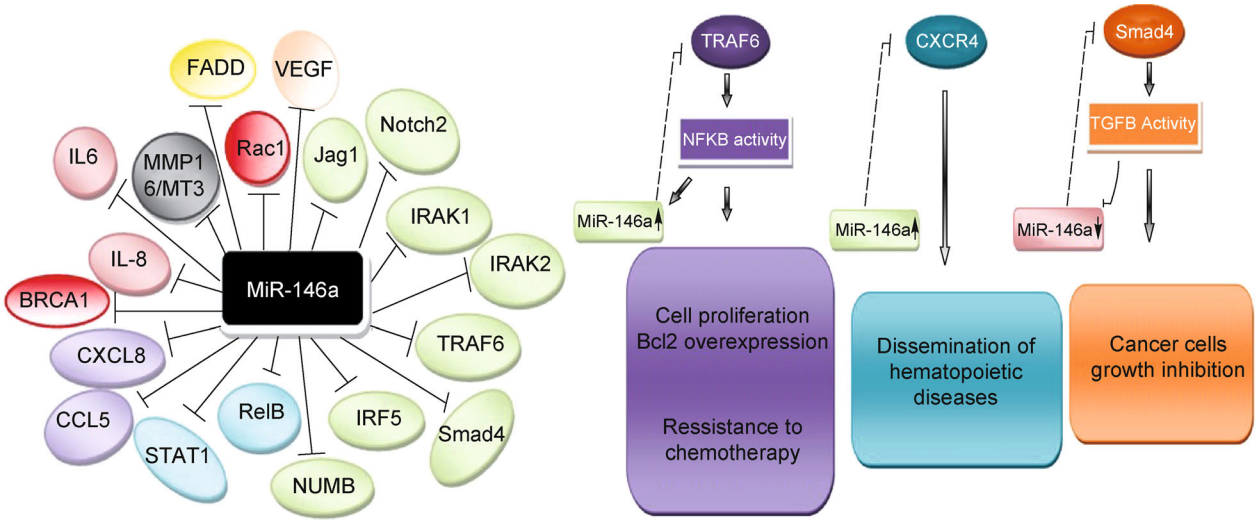

Figure 2 The role of miR-146a in targeting some of the leukemia genes. Abbreviations: FADD: Fas associated death domain protein, Rac1: ras-related C3 botulinum toxin substrate 1, VEGF: vascular endothelial growth factor, jag 1: jagged 1,IRAK1: interleukin-1 receptor-associated kinase 1, TRAF6: TNF-receptor-associated factor 4, IRF5: interferon regulatory factor 5, RelB: avian reticuloendotheliosis viral (v-rel) oncogene related B, STAT: signal transducer and activator of transcription 1, CCL5: chemokine (C-C motif) ligand 5, CXCL8: chemokine (C-X-C motif) ligand 8, BRCA1: breast cancer 1, IL: interleukin, MMP16: matrix metallopeptidase 16, MT3: metallothionein 3.

Acknowledgements

This paper is issued from thesis of Farzaneh Tavakoli, MSc student of hematology and blood banking. This work was financially supported by grant TH93/17 from vice chancellor for research affairs of Ahvaz Jundishapur University of Medical Sciences.

Conflict of interest statement

None of the authors of this paper have a financial or individualized relationship with other people or institutions that could inappropriately influence the content or directions of paper.

References

Béres N J, Szabó D, Kocsis D, Szűcs D, Kiss Z, Müller K E, Lendvai G, Kiss A, Arató A, Sziksz E, Vannay Á, Szabó A J, Veres G (2016). Role of Altered Expression of miR-146a, miR-155, and miR-122 in Pediatric Patients with Inflammatory Bowel Disease. Inflamm Bowel Dis, 22(2): 327–335

Bottoni A, Calin G (2013). A Calin G. MicroRNAs as main players in the pathogenesis of chronic lymphocytic leukemia. MicroRNA, 2(3): 158–164

Contreras J, Palanichamy J, Tran T, Fernando T, Rodriguez-Malave N, Goswami N, Arboleda V A, Casero D, Rao D S (2015). MicroRNA-146a modulates B-cell oncogenesis by regulating Egr1. Oncotarget, 6 (13): 11023–11037

Daschkey S, Röttgers S, Giri A,Bradtke J , Teigler-Schlegel A, Meister G, Borkhardt A, Landgraf P (2013). MicroRNAs distinguish cytogenetic subgroups in pediatric AML and contribute to complex regulatory networks in AML-relevant pathways. Plos One, 8(2): 276–276

de Oliveira J C, Scrideli C A, Brassesco M S, Morales A G, Pezuk J A, de Paula Queiroz R, Yunes J A, Brandalise S R, Tone L G(2012). Differential miRNA expression in childhood acute lymphoblastic leukemia and association with clinical and biological features. Leuk Res, 36(3): 293–298

Du J, Lu C, Cui G, Chen Y, He J (2013). DNA repair gene XRCC1 polymorphisms and susceptibility to childhood acute lymphoblastic leukemia: a meta-analysis. Chin J Cancer Res, 25(4): 405–415

Duyu M, Durmaz B, Gunduz C, Vergin C,Yilmaz Karapinar D, Aksoylar S, Kavakli K, Cetingul N, Irken G, Yaman Y, Ozkinay F, Cogulu O (2014). Prospective evaluation of whole genome microRNA expression profiling in childhood acute lymphoblastic leukemia. BioMed Research International, 2014(1):967585–967585

Ebert B L (2010). Genetic deletions in AML and MDS. Best Pract Res Clin Haematol, 23(4): 457–461

Fallah P, Arefian E, Naderi M, Aghaee-Bakhtiari S H, Atashi A, Ahmadi K, Shafiee A, Soleimani M (2013). miR-146a and miR-150 promote the differentiation of CD133 + cells into T-lymphoid lineage. Mol Biol Rep, 40(8): 4713–4719

Faraoni I, Antonetti F R, Cardone J, Bonmassar E (2009). miR-155 gene: a typical multifunctional microRNA. Biochimica et Biophysica Acta (BBA)-. Molecular Basis of Disease., 1792(6): 497–505

Ferreira A, Moura L, Tojal I, Ambrósio L, Pinto-Simões B, Hamerschlak N, Calin G A, Ivan C, Covas D T, Kashima S, Castro F A (2014). ApoptomiRs expression modulated by BCR–ABL is linked to CML progression and imatinib resistance. Blood Cells Mol Dis, 53(1): 47–55

Friedmann A M, Weinstein H J (2000). The role of prognostic features in the treatment of childhood acute lymphoblastic leukemia. Oncologist, 5(4): 321–328

Gaballa M R, Besa E C (2014). Myelodysplastic syndromes with 5q deletion: pathophysiology and role of lenalidomide. Ann Hematol, 93(5): 723–733

Ghose J, Bhattacharyya N P (2015). Transcriptional regulation of microRNA-100, −146a, and −150 genes by p53 and NFκB p65/RelA in mouse striatal STHdh(Q7)/ Hdh(Q7) cells and human cervical carcinoma HeLa cells. RNA Biol, 12(4): 457–477

Hagag A A, Nosair N A, Ghaith F M, Elshenawy E H (2014). Prognostic value of protease activated receptor-1 in children with acute lymphoblastic leukemia. Mediterr J Hematol Infect Dis, 6(1): e2014029

Han B W, Feng D D, Li Z G, Luo X Q, Zhang H, Li X J, Zhang X J, Zheng L L, Zeng C W, Lin K Y, Zhang P, Xu L, Chen Y Q (2011). A set of miRNAs that involve in the pathways of drug resistance and leukemic stem-cell differentiation is associated with the risk of relapse and glucocorticoid response in childhood ALL. Hum Mol Genet, 20(24): 4903–4915

Hauptman N, Glavac D (2013). MicroRNAs and long non-coding RNAs: prospects in diagnostics and therapy of cancer. Radiol Oncol, 47(4): 311–318

Jurado-Camino T, Córdoba R, Esteban-Burgos L, Hernández-Jiménez E, Toledano V, Hernandez-Rivas J A, Ruiz-Sainz E, Cobo T, Siliceo M, Perez de Diego R, Belda C, Cubillos-Zapata C, Lopez-Collazo E (2015). Chronic Lymphocytic Leukemia: A Paradigm of Innate Immune Cross-Tolerance. J Immunol, 194(2): 719–727

Li P, Wang X (2013). Role of signaling pathways and miRNAs in chronic lymphocytic leukemia. Chin Med J (Engl), 126(21): 4175–4182

Li X, Sanda T, Look A T, Novina C D, von Boehmer H (2011). Repression of tumor suppressor miR-451 is essential for NOTCH1-induced oncogenesis in T-ALL. J Exp Med, 208(4): 663–675

Mavrakis K J, Van Der Meulen J, Wolfe A L, Liu X, Mets E, Taghon T, Khan A A, Setty M, Rondou P, Vandenberghe P, Delabesse E, Benoit Y, Socci N B, Leslie C S, Van Vlierberghe P, Speleman F, Wendel H G (2011). A cooperative microRNA-tumor suppressor gene network in acute T-cell lymphoblastic leukemia (T-ALL). Nat Genet, 43(7): 673–678

Mei Y, Gao C, Wang K, Cui L, Li W, Zhao X, Liu F, Wu M, Deng G, Ding W, Jia H, Li Z (2014). Effect of microRNA‐210 on prognosis and response to chemotherapeutic drugs in pediatric acute lymphoblastic leukemia. Cancer Sci, 105(4): 463–472

Musilova K, Mraz M (2015). MicroRNAs in B-cell lymphomas: how a complex biology gets more complex. Leukemia, 29(5): 1004–1017

Naderi M, Abdul T H, Soleimani M, Shabani I, Hashemi S M (2015). A Home-brew Real-time PCR Assay for Reliable Detection and Quantification of Mature miR-122. Appl Immunohistochem Mol Morphol, 23(8):60–66

Portell C A, Wenzell C M, Advani A S (2013). Clinical and

pharmacologic aspects of blinatumomab in the treatment of B-cell acute lymphoblastic leukemia. Clin Pharmacol, 5: 5–11

Rosato P, Anastasiadou E, Garg N, Lenze D, Boccellato F, Vincenti S, Severa M, Coccia E M, Bigi R, Cirone M, Ferretti E, Campese A F, Hummel M, Frati L, Presutti C, Faggioni A, Trivedi P (2012). Differential regulation of miR-21 and miR-146a by Epstein–Barr virus-encoded EBNA2. Leukemia, 26(11): 2343–2352

Saki N, Abroun S, Soleimani M, Mortazavi Y, Kaviani S, Arefian E (2014). The roles of miR-146a in the differentiation of Jurkat T-lymphoblasts. Hematology., 19(3): 141–147

Schotte D, De Menezes R X, Moqadam F A, Khankahdani L M, Lange-Turenhout E,Chen C , Pieters P, Den Boer M L(2011) MicroRNA characterize genetic diversity and drug resistance in pediatric acute lymphoblastic leukemia. Haematologica, 96(5):703–711

Schotte D, Lange-Turenhout E A, Stumpel D J, Stam R W, Buijs-Gladdines J G, Meijerink J P, Pieters R, Den Boer M L (2010). Expression of miR-196b is not exclusively MLL-driven but especially linked to activation of HOXA genes in pediatric acute lymphoblastic leukemia. Haematologica. 95(10):1675–1682

So A Y L, Zhao J L, Baltimore D (2013). The Yin and Yang of microRNAs: leukemia and immunity. Immunol Rev, 253(1): 129–145

Starczynowski D T, Kuchenbauer F, Argiropoulos B, Sung S, Morin R, Muranyi A, Hirst M, Hogge D, Marra M, Wells R A, Buckstein R, Lam W, Humphries R K, Karsan A (2010). Identification of miR-145 and miR-146a as mediators of the 5q-syndrome phenotype. Nat Med, 16(1): 49–58

Visone R, Rassenti L Z, Veronese A, Taccioli C, Costinean S, Aguda B D, Volinia S, Ferracin M, Palatini J, Balatti V, Alder H, Negrini M, Kipps T J, Croce C M (2009). Karyotype-specific microRNA signature in chronic lymphocytic leukemia. Blood, 114(18): 3872–3879

Wang Y, Li Z, He C, Wang D, Yuan X, Chen J, Jin J (2010). MicroRNAs expression signatures are associated with lineage and survival in acute leukemias. Blood Cells Mol Dis, 44(3): 191–197

Xiong Q, Yang Y, Wang H, Li J, Wang S, Li Y, Yang Y, Cai K, Ruan X, Yan J, Hu S, Fang X (2014). Characterization of miRNomes in acute and chronic myeloid leukemia cell lines. Genomics Proteomics Bioinformatics, 12(2): 79–91

Yan J, Jiang N, Huang G, Tay J L S, Lin B, Bi C, Koh G S, Li Z, Tan J, Chung T H, Lu Y, Ariffin H, Kham S K Y, Yeoh A E J, Chng W J (2013). Deregulated MIR335 that targets MAPK1 is implicated in poor outcome of paediatric acute lymphoblastic leukaemia. Br J Haematol, 163(1): 93–103

Yin J Y, Tang Q, Qian W, Qian J, Lin J, Wen X M, Zhou J D, Zhang Y Y, Zhu X W, Deng Z Q (2014). Increased expression of miR-24 is associated with acute myeloid leukemia with t (8; 21). Int J Clin Exp Pathol, 7(11): 8032–8038

Zhang H, Luo X Q, Zhang P, Huang L B, Zheng Y S, Wu J, Zhou H, Qu L H, Xu L, Chen Y Q (2009). MicroRNA patterns associated with clinical prognostic parameters and CNS relapse prediction in pediatric acute leukemia. PLoS ONE, 4(11): e7826