

CLINICAL STUDY

MicroRNAs expression profiles as biomarkers and therapeutic tools in Turkish patients with chronic myeloid leukemia

Yurt M¹, Ayyildiz O², Karakus A², Nursal AF³, Isi H¹

Department of Medical Biology, Faculty of Medicine, Dicle University, Diyarbakir, Turkey.
feyda.nursal@gmail.com

ABSTRACT

AIM: In 95 % of Chronic myeloid leukemia (CML) patients, chromosomal translocation resulting in the formation of the Philadelphia (Ph) chromosome (t:9;22) is observed, which in turn leads to the formation of the *BCR-ABL* fusion gene. MicroRNAs (miRNAs) are a group of small and non-coding RNAs modulating gene expression via binding to the target mRNAs. We aimed to characterize the expression profiles of various miRNAs in different stages of Ph(+) CML patients.

METHODS: This case-controlled study was conducted in 75 CML patients and 25 healthy controls. The subjects were categorized into 4 groups; newly diagnosed patients, treatment-response patients, treatment-failure patients, and healthy controls. Expressions of miRNAs was analyzed by RT-PCR.

RESULTS: miR-150 expression was downregulated in the treatment failure patients compared to the control group ($p = 0.003212$) while miRNA 148b expression up-regulated in the treatment failure patients than the control group ($p = 0.038016$). miR-10a expression was up-regulated in newly diagnosed and treatment response patients compared to control group ($p = 0.003934$, $p = 0.000292$, respectively). It was found that miR-10a expression increased 11.17-fold in newly diagnosed patients and 9.82-fold in treatment response patients than in the control group.

CONCLUSION: Our data suggest that expression profiles of miR-10a, miR-150, and miRNA 148b were correlated as biomarker and therapeutic tool in Turkish patients with CML (Tab. 2, Fig. 1, Ref. 30). Text in PDF www.elis.sk.

KEY WORDS: chronic myeloid leukemia, microRNAs, new diagnosis, treatment response, treatment failure.

Introduction

Chronic myeloid leukemia (CML) is a clonal hematopoietic stem cell disease, manifested by an expansion of immature granulocytes (blasts) which amass in the bone marrow and hinder the normal blood cell production (1). Epidemiologic studies report that the frequency of CML is approximately 1–2 cases/100,000 people, and most CML cases are old people (median age on diagnosis: 65 years) (2). In 95 % of the cases, chromosomal translocation leading to the formation of the Philadelphia (Ph) chromosome is seen, which in turn causes the emergence of the *BCR-ABL* fusion gene (3). While the *BCR-ABL* fusion impels the initial chronic phase of CML, further genomic changes that render leukemia cells resistant to Tyrosine kinase inhibitors (TKIs) therapy and independent of *BCR-ABL* play a role in the progression of the disease.

MicroRNAs (*miRNAs*) are a class of small noncoding RNAs of 21–22 in length that play a role in the regulation of gene ex-

pression at the posttranscriptional level (4). They are evolutionarily well-conserved and, by binding to their target transcript in the 3'-UTR, can hinder the translation of proteins and destabilize the target mRNAs (5). Because of the regulation exerted by miRNAs on numerous biological processes, such as proliferation, apoptosis and differentiation, impairment of the miRNA regulatory network may be involved in tumorigenesis. miRNAs can serve as both oncogenes and tumor suppressors (6). miRNAs play role in CML pathogenesis: some miRNAs are up-regulated while others are down-regulated in the peripheral blood of CML patients (7). Furthermore, some studies suggest that different expression levels of miRNAs are seen in CML patients with good and poor response to TKI therapy.

Given that miRNAs have been implied to play a major role in leukemogenesis and that expression profiles of miRNAs have been linked to several cancer types, we aimed to test whether expression profiles of miR-150, miR-10a, miR-148b, and miR-130b display in patients at different stages of CML including newly diagnosed, treatment-response and therapy-failure patients.

Materials and methods*Study population*

This study was conducted from April 2017 to January 2018 and included 75 (32 males and 43 females) patients with CML

¹Department of Medical Biology, Faculty of Medicine, Dicle University, Diyarbakir, Turkey, ²Department of Hematology, Faculty of Medicine, Dicle University, Diyarbakir, Turkey, and ³Department of Medical Genetics, Faculty of Medicine, Hitit University, Corum, Turkey

Address for correspondence: A.F. Nursal, Department of Medical Genetics, Faculty of Medicine, Hitit University, Corum, Turkey.
Phone: +90.364.2221100, Fax: +90.364.2221102

recruited from the Hematology Department of Dicle University Research and Education Hospital, Diyarbakir, Turkey. An age- and gender-matched control group composed of 25 healthy subjects (11 males and 14 females) without a medical history of cancer or other chronic diseases. The diagnosis of CML was based on standard clinical data and confirmed by molecular analyses. The current CML treatments include hydroxyurea, bone marrow transplantation or TKIs. We separated all subjects into four groups: Group 1 (n = 25): newly diagnosed patients, Group 2 (n = 25): treatment response patients, Group 3 (n = 25): treatment failure patients, Group 4 (n = 25): healthy control group. Treatment response is characterized as $BCR-ABL < 0.1\%$. Therapy failure is defined here as complete hematological response with failure to achieve complete cytogenetic remission. All subjects signed a written consent form after being informed about the details of the study. Informed consent in accordance with the study protocol was approved by the Ethics Committee of the Dicle University. All the procedures were rigorously conducted following the terms of the Declaration of Helsinki.

Bcr-Abl analysis

Using RBC Lysis Solution and QiAmp RNA Blood Mini Kit kits, leukocytes from 10 ml whole blood were obtained first, then RNA was isolated from leukocytes. (Qiagen, Germantown, MD). Then, the total RNA quality was checked using the spectrophotometric method (Maestrogen, MaestroNano Spectrophotometer, USA). Then, the cDNAs were synthesized using a Ipsogen RT Kit (Qiagen Hilden, Germany). This was used for all samples to perform reverse transcription from 1 µg of total RNA in a Labcycler Thermal Cycler (SenSoquest). Following these procedures, the cDNA step was completed. Then for this step, the mix was loaded onto the Real-Time PCR (Rotor-Gene, Qiagen Hilden, Germany) using the Ipsogen BCR-ABL1 Mbc IS-MMR Kit. The RT-PCR step was switched to look at Bcr-Abl levels. According to the results, groups with newly diagnosed, treatment-responsive and unresponsive patients were formed.

RT-PCR analysis of plasma samples for miRNA levels

miRNA isolation was performed using miRNeasy Serum/Plasma Kit. The miRNAs obtained were stored at -80 °C until the Real-Time PCR (RT-PCR) stage. Then, the total RNA quality was checked using the spectrophotometric method (Maestrogen, MaestroNano Spectrophotometer, USA). Then, the cDNAs were synthesized using Qiagen miScript II Reverse Transcription Kit II (Qiagen, Sabioscience). This was used for all samples to perform reverse transcription from 0,5µg of miRNA in a Labcycler Thermal Cycler (SenSoquest). For RT-qPCR analysis, diluted cDNA was used as a template. The cDNA enrichment step proceeded then. cDNAs were enriched using the miScriptPreAMP PCR Kit. RT-PCR was introduced to determine the expression levels of miRNAs. The cDNA, in combination with a miScript SYBR Green PCR kit (Qiagen, Germantown, MD), was used for RT-qPCR miScriptPrimer Assays. RT-qPCR was run in a Rotor-Disc 72 with reaction volumes that used 25 µl of reaction mixture for 40 cycles of 95 °C for 15 min, 94 °C for 15 s, 55 °C for 30 s in

Tab. 1. Mean ages of the subjects.

Groups	Males		Females	
	n (%)	Mean age	n (%)	Mean age
Group 1	10 (10)	61.00±18.42	15 (15)	41.00±10.21
Group 2	10 (10)	48.60±18.57	15 (15)	48.87±11.54
Group 3	12 (12)	51.25±17.94	13 (13)	49.00±16.66
Group 4	11 (11)	53.36±12.64	14 (14)	47.64±16.64

Group 1: newly diagnosed patients; Group 2: treatment response patients; Group 3: treatment failure patients; Group 4: healthy controls.

the Qiagen Rotorgene Q (Qiagen, Hilden). The reaction mixture contained 10 µL SYBR Green Master, 2 µL miscript primer assay, 2 µL universal primer 1 µL of DNase/RNase-free distilled water and 5 µL of cDNA template. It is known that Snord61 is the most stable reference gene under exposure to pesticide, so we selected this gene in our study. Reference sequence numbers for all primers were obtained from the set shown in Table 1. (Genes were produced for Human (*Homo sapiens*) by miScript Primer Assays (Qiagen SABiosciences USA).

Ct values were exported to an Excel file, a table of Ct values was created, and the table was uploaded to the data analysis web tool (8). The samples were classified and tagged as the control and the patient groups. Manually selected reference genes were used to normalize the obtained Ct values. The online tool uses the $\Delta\Delta Ct$ method to calculate the fold-change/regulation. Briefly, ΔCt was first calculated between the studied gene of interest (GOI) and an average of housekeeping genes (HKG), and $\Delta\Delta Ct$ was further determined by subtracting the ΔCt (control) value from the ΔCt (experiment). Fold-change was assessed using the formula: $2^{(-\Delta\Delta Ct)}$. The data analysis report was exported from QIAGEN web portal (8).

Fold-change

Fold-change ($2^{(-\Delta\Delta Ct)}$) is the ratio of the normalized gene expression ($2^{(-\Delta Ct)}$) in the test sample over the normalized gene expression ($2^{(-\Delta Ct)}$) in the control sample. Fold-regulation, on the other hand, is a representation of the fold-change results in a biologically meaningful manner. In brief, fold-regulation is equal to fold-change in cases of positive- or up-regulation, where the fold-change value is greater than one. Conversely, when the fold-change is less than one, which is indicative of a negative- or down-regulation, fold-regulation equals to $(-1/(\text{fold-change}))$. Student's t-test was used to assess the relationship between the replicate $2^{(-\Delta Ct)}$ values obtained from each gene in the control group and those from the patient group, and the p values were calculated.

Statistical analysis

Real-time PCR data were analyzed using the Gene Globe Data Analysis Center (Qiagen). The raw data based on the $\Delta\Delta Ct$ method were normalized using the Snord61 housekeeping gene. Fold-change values ≤ 2 were considered to be up- or down-regulated. Comparison of continuous data was performed using Student's t-test, either paired or unpaired, as needed. p values less than 0.05 were deemed statistically significant.

Tab. 2. Fold change values according to groups.

Genes	Fold-Changes	p
Group 1		
Snord61	1.00	Housekeeping gene
miR-150	0.98	0.863306
miR-10a	11.17	0.003934
miR-148b	3.78	0.125377
miR-130b	1.08	0.617569
Group 2		
Snord61	1.00	Housekeeping gene
miR-150	1.58	0.237789
miR-10a	9.82	0.000292
miR-148b	1.97	0.451892
miR-130b	1.03	0.271973
Group 3		
Snord61	1.00	Housekeeping gene
miR-150	0.08	0.003212
miR-10a	2.85	0.375747
miR-148b	0.35	0.038016
miR-130b	0.77	0.105655
Group 4		
Snord61	1.00	Housekeeping gene
miR-150	0.05	0.000001
miR-10a	0.29	0.006950
miR-148b	0.18	0.003557
miR-130b	0.75	0.439569

Group 1: newly diagnosed; Group 2: treatment response; Group 3: treatment failure; Group 4: healthy control.

Results

A total of 100 subjects including 75 CML patients and 25 unrelated healthy subjects were evaluated. There were 43 % females and 32 % males in patients group. The mean age in the groups is shown in Table 1. To decipher a potential miRNA expression signature, we have analyzed expression profiles of miRNAs of newly diagnosed patients, treatment response patients, treatment failure patients, and compared them with healthy controls. The expression profiles of the groups are summarized in Table 2.

miR-150 expression was down-regulated in the treatment failure patients compared to the control group (p = 0.003212) while

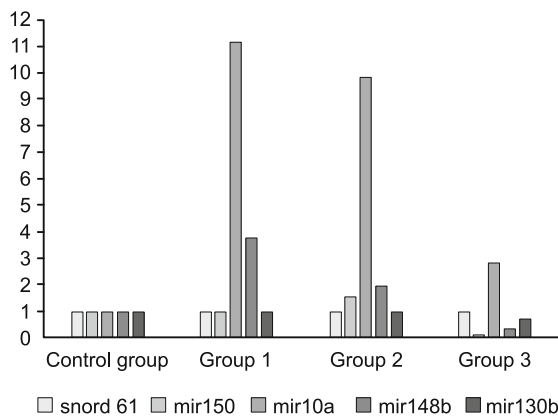


Fig. 1. Graphic view of the fold change of expression profiles of different miRNAs in groups (Group 1: newly diagnosed; Group 2: treatment response; Group 3: treatment failure and control group).

miRNA 148b expression was up-regulated in the treatment failure patients compared to the control group (p = 0.038016).

miR-10a expressions were up-regulated (fold change > 1) in newly diagnosed and treatment response patients compared to healthy control group (p = 0.003934, p = 0.000292, respectively). It was found that miR-10a expression increased 11.17-fold in newly diagnosed patients and 9.82-fold in the treatment response patients compared to the healthy control group. Although it was not statistically significant, miR-10a expression was 2.85 fold higher in the treatment failure patients compared to the healthy control group (fold change > 1) (p = 0.375747).

miR-130b expression differences were not statistically significant in all groups according to fold changes.

Graphic view of the fold changes of expression profiles of different miRNAs in groups is presented in Figure 1.

Discussion

CML is a group of cancers that are manifested by a unique genetic distortion, the breakpoint cluster region-Abelson murine leukemia viral oncogene homolog 1 (BCR-ABL1) fusion gene (9). Owing to the fact that this molecular lesion leads to CML, it was possible to create an effective targeted therapy that selectively hinders the tyrosine kinase activity of distorted BCR-ABL protein. Imatinib (IM) is a targeted therapy drug that can recognize and inactivate tyrosine kinase activity of the BCR-ABL oncoprotein, thus blocking leukemogenesis. Treatment of CML patients with IM produced very good responses, with regard to symptom management and hematological parameters. On the other hand, due to constant IM presence, primary and secondary resistance as well as molecular evidence of persistent malignancy have been observed in many CML patients (10). In some cases, point mutations in the BCR-ABL kinase domain have been increased in the development of IM resistance, secondary resistance particularly, eventually causing treatment failure. Resistance and intolerance to treatment remain a major clinical problem.

miRNAs are short, noncoding RNAs that are involved in post-transcriptional modulation of messenger RNAs (mRNAs). The human genome contains approximately 2 000 distinct miRNAs (11). The miRNAs play crucial roles in tumorigenesis and cancer progression via regulation of neoplasia onset, growth, metastasis, and resistance to treatment (12), in agreement with the role of miRNAs in several developmental processes, their misregulation can largely promote the phenotypic features of all cancer subtypes studied so far. The most common deregulated miRNAs in CML include miR-10a, miR-17/92, miR-150, miR-203, and miR-328 (13). In CML, enhanced expression of *miR-150* and *miR-146a*, and decreased expression of *miR-142-3p* and *miR-199b-5p* were seen after 15 days of TKI therapy (14), indicating that this drug has the capacity to rearrange the miRNA profiles of tumor cells.

In this study, we analyzed 75 BCR-ABL positive patients. Our goal was to identify distinctive miRNA signatures for CML subgroups. *miR-150* has invariably appeared to be down-regulated across multiple studies suggesting it as a potential candidate for early CML diagnosis. Multiple reports imply that reduced expres-

sion of *miR-150* represents poor prognosis and a more severe state of CML, whereas it was reported that reintroduction of *miR-150* relieved symptoms in cell lines (15). Down-regulation of *miR-150* was seen in CD34⁺ cells derived from six chronic phase CML patient samples (15), indicating that the down-regulation of this miRNA is involved in disease onset. Moreover, evidence for down-regulation of *miR-150* as a diagnostic biomarker of CML was demonstrated in a study that used a RT-PCR method in 50 newly diagnosed chronic phase CML patients, and found important down-regulation of *miR-150* (16). A study in 2010 conducted a microarray analysis in 10 chronic phase CML patients and validated the potency of *miR-150* as a biomarker for CML diagnosis (14). *miR-150* expression levels can also be helpful as a biomarker for treatment response. The down-regulation of *miR-150* was found in the BCR-ABL transformed leukemia cell line Mo7e-p210 (megakaryoblast), and could be reestablished in response to IM treatment (15). In this study, we found that miR-150 expression was down-regulated in patients with treatment failure compared to healthy controls ($p=0.003212$). Our results were compatible with those found in other studies.

There is growing evidence suggesting that miR-10a/b may serve as new oncogene in several types of human cancer, such as metastatic breast cancer, pancreatic cancer, esophageal cancer, hepatocellular carcinoma, nasopharyngeal carcinoma and colorectal cancer (17). These findings advocate that miR-10a and miR-10b are vigorously expressed in highly metastatic cancer and play significant roles in metastasis. *miR-10a* is a potential candidate for CML diagnosis. Using a Q-RT PCR method in 85 newly diagnosed chronic phase CML patients, down-regulation of *miR-10a* was seen in 71 % of patients, exhibiting its clinical pertinence as a biomarker for diagnosis (15). Other studies have also investigated the pertinence of *miR-10a* as a biomarker for drug response. *miR-10a* levels in the Mo7e-p210 cell line were unaffected by IM treatment (14), nevertheless, a significant increment of *miR-10a* was seen in microarray analysis of patient samples 15 days after IM treatment (14). In the present study, we found that miR-10a expression was up-regulated 11.17-fold in newly diagnosed and 9.82-fold in treatment response patient groups ($p = 0.003934$, $p = 0.000292$, respectively) (Tab. 2). It was thought that miR-10a expression was associated with treatment response.

miR-148a, miR-148b, and miR-152 are three members of the miR-148/152 family. They have similar sequences, configuration, and the same seed region (18). All of them have been reported to be down-regulated/up-regulated in numerous distinct types of tumors (e.g., gastrointestinal, ovarian, hepatocellular carcinoma, and pancreatic cancer) (19). Some studies have shown that members of the miR-148/152 family are expressed differentially in hematological malignancies including acute lymphoblastic leukemia (ALL), multiple myeloma (MM), and lymphoma (20–23). Huang et al. found that six miRNAs such as miR-148a were significantly up-regulated in the plasma of MM patients and high levels of miR-148a were associated with shorter relapse-free survival times (22). Wang et al. found a significantly lower expression of down-regulated miR-148/152 in AML patients compared to healthy controls (24). It was reported that low expression of miR-148b was found in a subset of CML patients with stable complete molecular responses

after stopping IM treatment (25). In this study, it was detected that miR-148b expression was up-regulated in treatment-failure patient group compared to the control group ($p = 0.038016$).

miR-130a plays distinct roles as oncogene or tumor suppressor gene to mediate diverse biological processes by modulating several canonical pathways or target genes in various cancer types. It has been found that the expression of miR-130a is distorted in some types of cancer, overexpressed in adult T cell leukemia (ATL) (26), gastric cancer (27), but down-expressed in bladder cancer (28), and chronic lymphocytic leukemia (29). Ding et al identified miR-130a as significantly over-expressed in t(8;21) AML (30). We found miR-130b expression showed no significant differences in all groups ($p > 0.05$).

Conclusion

In summary, our data demonstrated that miR-150, miR-10a, and miR-148b are deregulated at different stages of CML. The results of this study summarize the mechanisms by which miRNAs may play a role in CML pathogenesis. However, if they act in BCR-ABL dependent or independent manner has to be further investigated.

References

1. Jurkovicova D, Lukackova R, Magyerkova M et al. microRNA expression profiling as supportive diagnostic and therapy prediction tool in chronic myeloid leukemia. *Neoplasma* 2015; 62 (6): 949–958.
2. Hehlmann R, Hochhaus A, Baccarani M. European LeukemiaNet: Chronic myeloid leukaemia. *Lancet* 2007; 370 (9584): 342–350.
3. Kalidas M, Kantarjian H, Talpaz M. Chronic myelogenous leukemia. *JAMA* 2001; 286 (8): 895–898.
4. Lagos-Quintana M, Rauhut R, Lendeckel W, Tuschl T. Identification of novel genes coding for small expressed RNAs. *Science* 2001; 294 (5543): 853–858.
5. Baek D, Villen J, Shin C, Camargo FD, Gygi SP, Bartel DP. The impact of microRNAs on protein output. *Nature* 2008; 455 (7209): 64–71.
6. Garzon R, Calin GA, Croce CM. MicroRNAs in cancer. *Annu Rev Med* 2009; 60:167–179.
7. Zhao H, Wang D, Du W, Gu D, Yang R. MicroRNA and leukemia: tiny molecule, great function. *Crit Rev Oncol Hematol* 2010; 74 (3): 149–155.
8. <http://www.qiagen.com/geneglobe>.
9. Sun QC, Liu MB, Shen HJ et al. Inhibition by imatinib of expression of O-glycan-related glycosyltransferases and tumor-associated carbohydrate antigens in the K562 human leukemia cell line. *Asian Pac J Cancer Prev* 2013; 14 (4): 2447–2451.
10. Jiye A, Qian S, Wang G et al. Chronic Myeloid Leukemia Patients Sensitive and Resistant to Imatinib Treatment Show Different Metabolic Responses. *PLoS One* 2010; 5 (10): e13186.
11. Friedländer MR, Lizano E, Houben AJ et al. Evidence for the biogenesis of more than 1,000 novel human microRNAs. *Genome Biol* 2014; 15 (4): R57.
12. Xue J, Niu J, Wu J, Wu ZH. MicroRNAs in cancer therapeutic response: friend and foe. *World J Clin Oncol* 2014; 5 (4): 730–743.

13. **Gordon JE, Wong JJ, Rasko JE.** MicroRNAs in myeloid malignancies. *Br J Haematol* 2013; 162 (2): 162–176.
14. **Flamant S, Ritchie W, Guilhot J et al.** Micro-RNA response to imatinib mesylate in patients with chronic myeloid leukemia. *Haematologica* 2010; 95 (8): 1325–1333.
15. **Agirre X, Jiménez-Velasco A, San José-Enériz E et al.** Down-regulation of hsa-miR-10a in chronic myeloid leukemia CD34+ cells increases USF2-mediated cell growth. *Mol Cancer Res* 2008; 6 (12): 1830–1840.
16. **Fallah P, Amirzadeh N, Poopak B et al.** Expression pattern of key microRNAs in patients with newly diagnosed chronic myeloid leukemia in chronic phase. *Int J Lab Hematol* 2015; 37 (4): 560–568.
17. **Bi L, Sun L, Jin Z, Zhang S, Shen Z.** MicroRNA-10a/b are regulators of myeloid differentiation and acute myeloid leukemia. *Oncol Lett* 2018; 15 (4): 5611–5619.
18. **Calin GA, Sevignani C, Dumitru CD et al.** Human microRNA genes are frequently located at fragile sites and genomic regions involved in cancers. *Proc Natl Acad Sci USA* 2004; 101 (9): 2999–3004.
19. **Chen Y, Song YX, Wang ZN.** The MicroRNA-148/152 Family: Multifaceted Players. *Mol Cancer* 2013; 12: 43.
20. **Rio-Machin A, Ferreira BI, Henry T et al.** Downregulation of specific miRNAs in hyperdiploid multiple myeloma mimics the oncogenic effect of IgH translocations occurring in the non-hyperdiploid subtype. *Leukemia* 2013; 27 (4): 925–931.
21. **Wu Y, Liu GL, Liu SH, Wang CX, Xu YL, Mao P.** MicroRNA-148b enhances the radiosensitivity of non-Hodgkin's Lymphoma cells by promoting radiation-induced apoptosis. *J Radiat Res* 2012; 53 (4): 516–525.
22. **Huang JJ, Yu J, Li JY, RQ Z.** Circulating microRNA expression is associated with genetic subtype and survival of multiple myeloma. *Med Oncol* 2012; 29 (4): 2402–2408.
23. **Stumpel DJ, Schotte D, Lange-Turenhout EA et al.** Hypermethylation of specific microRNA genes in MLL-rearranged infant acute lymphoblastic leukemia: Major matters at a micro scale. *Leukemia* 2011; 25 (3): 429–439.
24. **Wang XX, Zhang R, Li Y.** Expression of the miR-148/152 Family in Acute Myeloid Leukemia and its Clinical Significance. *Med Sci Monit* 2017; 23: 4768–4778.
25. **Ohyashiki JH, Ohtsuki K, Mizoguchi I et al.** Downregulated microRNA-148b in circulating PBMCs in chronic myeloid leukemia patients with undetectable minimal residual disease: a possible biomarker to discontinue imatinib safely. *Drug Des Devel Ther* 2014; 8: 1151–1159.
26. **Ishihara K, Sasaki D, Tsuruda K et al.** Impact of miR-155 and miR-126 as novel biomarkers on the assessment of disease progression and prognosis in adult T-cell leukemia. *Cancer Epidemiol* 2012; 36 (6): 560–565.
27. **Jiang H, Yu WW, Wang LL, Peng Y.** MiR-130a acts as a potential diagnostic biomarker and promotes gastric cancer migration, invasion and proliferation by targeting RUNX3. *Oncol Rep* 2015; 34 (3): 1153–1161.
28. **Ratert N, Meyer HA, Jung M.** miRNA profiling identifies candidate mirnas for bladder cancer diagnosis and clinical outcome. *J Mol Diagn* 2013; 15 (5): 695–705.
29. **Kovaleva V, Mora R, Park YJ et al.** miRNA-130a targets ATG2B and DICER1 to inhibit autophagy and trigger killing of chronic lymphocytic leukemia cells. *Cancer Res* 2012; 72 (7): 1763–1772.
30. **Ding C, Chen SN, Macleod RAF et al.** MiR-130a is aberrantly over-expressed in adult acute myeloid leukemia with t(8; 21) and its suppression induces AML cell death. *Ups J Med Sci* 2018; 123 (1): 19–27.

Received June 2, 2019-.
Accepted October 14, 2019.