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LncRNA-H19 inhibits apoptosis of acute myeloid leukemia cells *via* targeting miR-29a-3p

T.-T. ZHAO[1], X. LIU[2]

[1]Department of Laboratory Medicine, Affiliated Municipal Hospital of Xuzhou Medical University, Xuzhou, China
[2]Department of Hematology, The Affiliated Hospital of Liaoning University of Traditional Chinese Medicine, Shenyang, China

Abstract. – OBJECTIVE: To explore the influences of long non-coding ribonucleic acid (lncRNA)-H19 on the proliferation and apoptosis of acute myeloid leukemia (AML) cells *via* the Wnt signaling pathway. PATIENTS AND METHODS: Blood samples were collected from 40 AML patients. The AML cells were cultured. Cell counting kit-8 (CCK-8) was used to detect cell proliferation and flow cytometry was applied to analyze cell cycle and determine the apoptosis rate. Moreover, the action target of lncRNA-H19 was detected through a dual-luciferase reporter assay and Western blotting was performed to detect the change in protein level.

RESULTS: The expression of lncRNA-H19 in AML patients was markedly higher than that in normal controls and compared with human embryonic kidney (HEK)-293T cells, AML cell Kasumi-1 exhibited an increased lncRNA-H19 expression. LncRNA-H19 could promote cell proliferation, but suppress cell apoptosis. It is bound to micro RNA (miR)-29a-3p in a targeted manner. and the expression level of miR-29a-3p in AML patients was prominently lower than that in normal controls. After miR-29a-3p was inhibited, the expression of intranuclear β-catenin was significantly increased and the Wnt/β-catenin pathway critical molecules T-cell factor (TCF) and lymphoid enhancer factor 1 (LEF1) were evidently up-regulated after the down-regulation of miR-29a-3p.

CONCLUSIONS: LncRNA-H19 targets miR-29a-3p to promote the proliferation of AML cells, but inhibit the apoptosis through the Wnt/ β-catenin signaling pathway.

Key Words:
LncRNA-H19, Wnt/β-catenin signaling pathway, Acute myeloid leukemia, Proliferation apoptosis.

Introduction

Acute myeloid leukemia (AML) is one of the most common hematological malignancies. According to the SEER database and epidemiological statistics, there are about 20,830 new cases of AML every year in America, accounting for 1.3% of the total[1]. In addition, the number of people who die of AML reaches about 10,460, which is 1.8% of the total cancer deaths. AML cases account for about 70% of all the patients with acute leukemia[2,3]. What's worse, the AML is rising year by year due to industrial and agricultural pollution. Currently, numerous reports[4-7] have suggested that epigenetic changes such as micro ribonucleic acid (miRNA) expression abnormality, deoxyribonucleic acid (DNA) and RNA methylation changes, as well as histone modification disorder play important roles in modulating the occurrence and development of AML. Therefore, it is particularly important to explore the epigenetics and pathogenesis of AML to raise the efficacy in the treatment of AML.

Long non-coding RNAs (lncRNAs) are considered as non-protein-coding RNAs with the length of more than 200 nt. They used to be regarded as the "noises" in genomes, while they have recently been found to participate in many biological activities, such as cell growth, proliferation, invasion, and apoptosis. LncRNA-H19 is a ncRNA measuring 3,000 bp in length and located on chromosome 11p15.5, it is expressed in the nucleus and cytoplasm[8,9]. LncRNA-H19 acts as an oncogene to take part in various pathological processes in the growth and metastasis of tumors, including breast cancer, bladder cancer, colorectal cancer, gastric cancer, head, and neck squamous cell carcinoma and esophagus cancer[10-12]. In these tumor tissues, the lncRNA-H19 expression is significantly elevated and the over-expression of lncRNA-H19 can accelerate cancer cell proliferation, migration, invasion, and metastasis.

As endogenous ncRNAs, miRNAs modulate the gene expression through the degradation of

Corresponding Author: Xin Liu, MM; e-mail: liuxin19730512@163.com

Table I. Primer sequences.

Gene	Forward	Reverse
LncRNA-H19	5'-ATGGTGCTACCCAGCTCAAG-3'	5'-GCTGTTCCGATGGTGTCTTT-3'
MiR-29a-3p	5'-CTGGTGTCGTGGAATTCAGTTGA-3'	5'-CCTGGCTCCTCACTTGGC-3'
β-actin	5'-AAGTACTCCGTGTGGATCGG-3'	5'-ATGCTATCACCTCCCCTGTG-3'

mRNAs and the inhibition on their post-transcriptional translation[13]. According to the findings in several studies[14], lncRNAs, as endogenous RNAs (ceRNAs) competing with miRNAs, affect the expression of miRNA targets. However, there have been no reports on whether lncRNA-H19 acts as a ceRNA to regulate the expression of target protein-binding miRNA in AML now.

In the present work, it was assumed that lncRNA may suppress the expression of miR-29a-3p to promote the proliferation of AML cells. In the preliminary research, the patients with differentially expressed lncRNAs were first screened from the AML samples, while the expressions of lncRNA-H19 and miR-29a-3p in AML patients were detected using the Polymerase Chain Reaction (PCR) chip. The potential mechanism was further analyzed by which lncRNA-H19 promotes the occurrence and development of AML *in vitro*. The present study can help to better understand the pathogenesis of AML and provides a potential target for the intervention and treatment of AML.

Patients and Methods

Clinical Samples

Blood samples were collected from 40 AML patients who were hospitalized from January 2018 to December 2018 in the Department of Hematology of our hospital. All the patients were informed of this research which was approved by the Ethics Review Committee of the hospital.

Cell Culture

AML cell line Kasumi-1 and human embryonic kidney (HEK)-293T cell line were purchased from the Shanghai Cell Bank of Chinese Academy of Sciences (Shanghai, China) and they were cultured in a humid Roswell Park Memorial Institute-1640 (RPMI-1640) medium (HyClone; South Logan, UT, USA) containing 10% fetal bovine serum (FBS; Gibco, Rockville, MD, USA) at 37°C. The medium was replaced once every other day and the cells were subcultured when the fusion rate reached 80-90%.

Reverse Transcription (RT)-PCR

Total RNAs were extracted from all blood and cells using the TRIzol reagent (Invitrogen, Carlsbad, CA, USA). Complementary DNAs (cDNAs) were synthesized using RT^2 First Strand Kit (Qiagen, Hilden, Germany) and NCode^{TM} miRNA Fist-Strand cDNA synthesis kit (Life Technologies, Carlsbad, CA, USA) separately. RT-PCR was performed in the ABI Q6 detection system (Applied Biosystems, Foster City, CA, USA) using the SYBR real-time master mix kit (cat No.: 330401). The following specific primers were used for the PCR (Table I).

Cell Proliferation Assay

According to the manufacturer's instructions, the proliferation of cells was determined *via* cell counting kit-8 (CCK-8; Dojindo Laboratories, Kumamoto, Japan). The cells were inoculated in a 96-well plate at an initial density of 2×10^4 cells/mL and added with the CCK-8 solution (10 μL/well) at 12, 24, 48, or 72 h to measure cell viability. After they were incubated with 5% CO_2 at 37°C for 2 h, the absorbance of each well was measured at 450 nm.

Detection of Apoptosis Using Flow Cytometry

The cells differently treated were collected and cultured independently. Each group of Kasumi-1 cells was collected in accordance with the instructions in the flow cytometer kit and washed using phosphate-buffered saline (PBS; Beoytime, Shanghai, China). After digestion, the cell suspension was prepared at a concentration of 1×10^6 cells/mL and then centrifuged. Next, the supernatant was discarded. Subsequently, the cells were added with 5 μL Annexin V-FITC and 10μL propidium iodide (PI) and slightly mixed, followed by incubation for 5-15 min. The apoptosis rate was determined using the flow cytometer and

cell sorter system. The flow cytometer (FACScan, BD Biosciences, San Diego, CA, USA) equipped with CellQuest software (BD Biosciences, San Diego, CA, USA) was adopted to analyze and differentiate the cells: living cells, dead cells, and apoptotic cells at early and advanced stages. In each experiment, the percentages of the apoptotic cells at early and advanced stages were compared with those in control group, respectively. The percentages were determined for 3 times.

Western Blotting

Proteins were extracted using radioimmunoprecipitation assay (RIPA) buffer (Beoytime, Shanghai, China), isolated *via* sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) and electro transferred onto a polyvinylidene difluoride membrane (PVDF; Millipore, Billerica, MA, USA). After a successful protein transfer, the membrane was sealed in 5% skim milk at 37°C for 2 h and incubated with the primary antibody at 4°C overnight, followed by incubation with the horseradish peroxidase (HRP)-linked secondary antibody at 37°C for 1 h. The bands were observed using an enhanced chemiluminescence assay (ECL) kit as specified by the manufacturers.

Statistical Analysis

Statistical Product and Service Solutions (SPSS) 22.0 (IBM, Armonk, NY, USA) and GraphPad Prism 5.0 (GraphPad, La Jolla, CA, USA) were applied for statistical analysis. All results were expressed as mean ± standard deviation (SD) and the data were analyzed using the two-tail *t*-test, one-way analysis of variance (ANOVA) and chi-square test. $p<0.05$ suggested that the difference was statistically significant.

Results

LncRNA-H19 Was Highly Expressed in AML Patients and Cells

The expression of lncRNA-H19 was detected using qRT-PCR among 40 AML patients and AML cell line Kasumi-1. According to the results, AML patients exhibited a significantly higher expression level of lncRNA-H19 than normal controls ($p<0.01$) (Figure 1A). Compared with that in HEK-293T cells, the expression level of lncRNA-H19 was increased ($p<0.01$) (Figure 1B), suggesting that lncRNA-H19 may be a potential risk factor for AML.

LncRNA-H19 Promoted the Proliferation of AML Cells

The cells were transfected with small-interfering (si)-H19 and H19-OE separately, and the proliferation capacity was determined. It was found that the proliferation capacity of the cells transfected with si-H19 was evidently weakened, but those transfected with H19-OE had a markedly enhanced proliferation capacity ($p<0.001$) (Figure 2A). The cell cycle was further analyzed and it was discovered that H19-OE blocked Gap

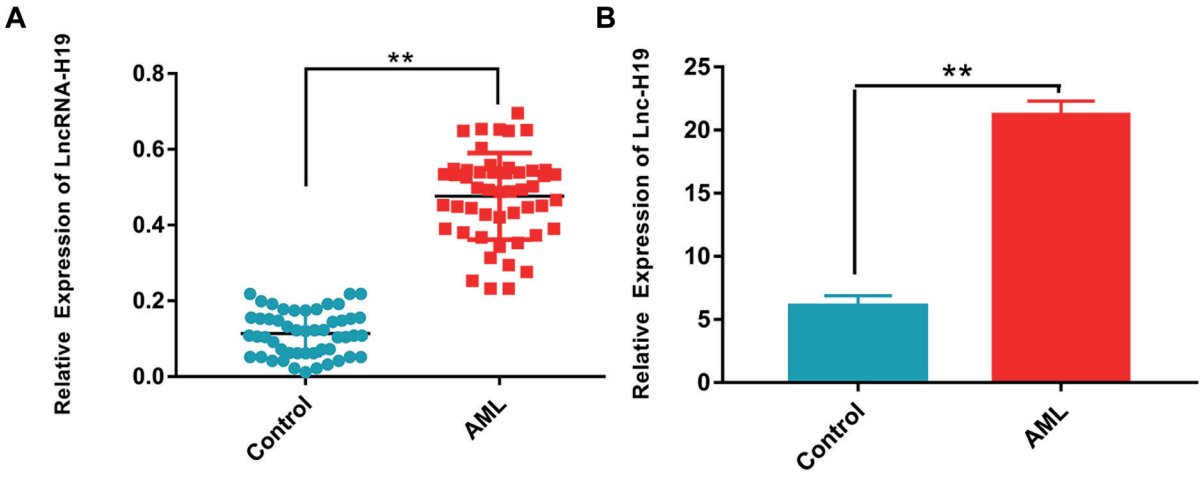

Figure 1. ***A,*** Expression of serum lncRNA-H19 in 40 AML patients and normal controls detected *via* qRT-PCR. ***B,*** Expression of lncRNA-H19 in AML cell line Kasumi-1 detected *via* qRT-PCR.

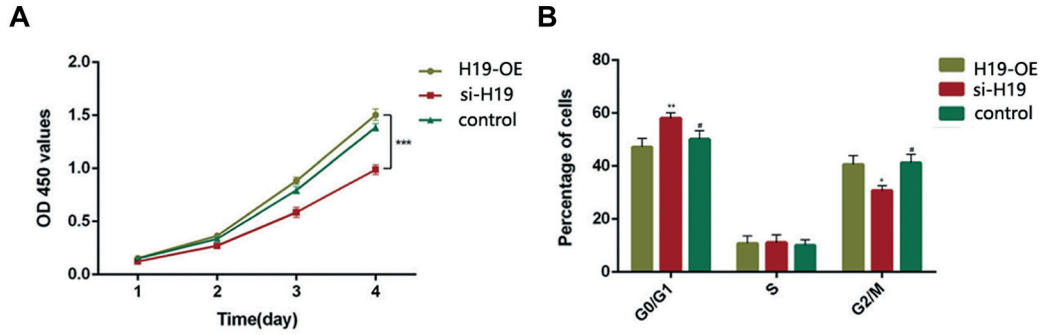

Figure 2. ***A,*** Cell proliferation capacity detected: si-H19-transfected cells exhibit a markedly weakened proliferation capacity, but the proliferation capacity of the H-19-OE-transfected cells is notably strengthened (p<0.001). ***B,*** H19-OE blocks G0/G1 (p<0.001).

0 (G0)/G1 (p<0.001) (Figure 2B). These results indicate that lncRNA-H19 can accelerate cell proliferation.

LncRNA-H19 Suppressed the Apoptosis of AML Cells

After lncRNA-H19 was over-expressed or its expression was inhibited, the cell apoptosis rate was determined using the flow cytometer. According to the results, the cell apoptosis rates in H19-OE, si-H19, and control groups were (5.21±0.22)%, (31.3±11.34)%, and (15.21±2.22)%, respectively, displaying statistical differences (p<0.05) (Figure 3).

LncRNA-H19 Targeted to Bind to MiR-29a-3p According to the Luciferase Reporter Assay

The target of lncRNA-H19 was predicted by a bioinformatic method and the results showed that after the over-expression of lncRNA-H19, the expression of miR-29a-2p was lowered, and the Wnt pathway was activated (Figure 4A). The further prediction results revealed that lncRNA-H19 contained a complementary site to the 3'-UTR of miR-29a-3p (Figure 4B).

The binding site of the mutant lncRNA-H19 was detected. It was discovered through the luciferase reporter assay that only after the interaction between the wild-type lncRNA-H19 and miR-29a-3p, the fluorescence intensity was significantly weakened (p<0.05) (Figure 5A). The expression of miR-29a-3p in AML patients was further analyzed and the results displayed that the expression level of miR-29a-3p in AML patients was markedly lower than that in normal controls (p<0.01) (Figure 5B).

MiR-29a-3p Targeted and Modulated the Wnt/β-Catenin Pathway

To further verify whether miR-29a-3p targets the Wnt/β-catenin pathway, the qRT-PCR and WB were conducted. The analysis results

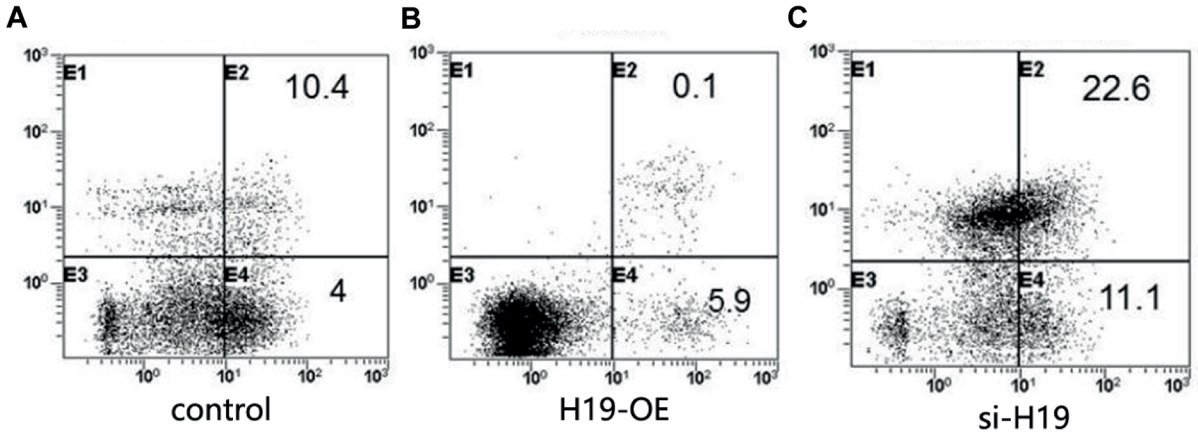

Figure 3. Cell apoptosis rate determined using flow cytometry. Cell apoptosis rates in H19-OE, si-H19, and control groups are (5.21 ±0.22)%, (31.3 ± 11.34)%, and (15.21±2.22)%, respectively (p<0.05).

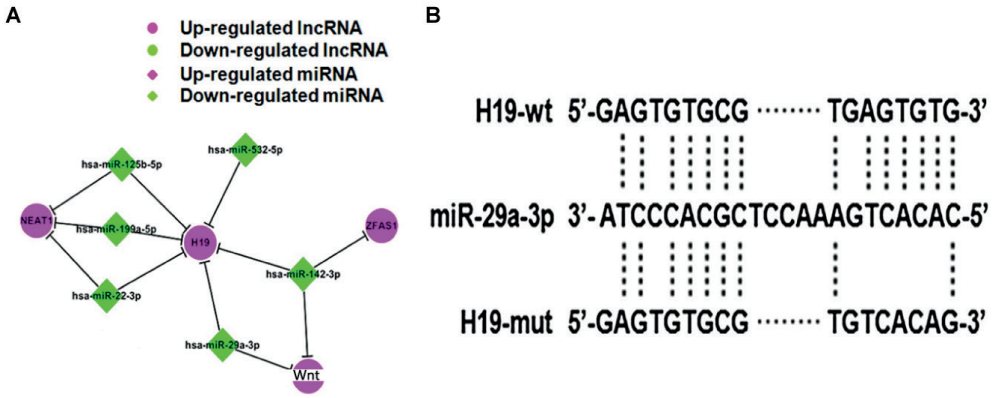

Figure 4. ***A,*** After the over-expression of lncRNA-h19, the expression of miR-29a-2p is lowered, and the Wnt pathway is activated. ***B,*** LncRNA-H19 contains a complementary site to the 3'-UTR of miR-29a-3p.

showed that the expression of β-catenin in the nuclei was significantly increased after miR-29a-3p was inhibited (Figure 6A) ($p<0.05$), and that the Wnt/β-catenin pathway critical molecules T-cell factor (TCF) and lymphoid enhancer factor 1 (LEF1) were evidently up-regulated after miR-29a-3p was down-regulated (Figure 6B) ($p<0.05$).

Discussion

AML is a malignant hematopathy with high heterogeneity, in which bone marrow progenitors are stopped from differentiating at different stages, leukemia cell proliferation is out of control, and cell apoptosis is impaired, thereby inhibiting the hematopoietic system, and its complex pathogenesis has not yet been fully elucidated. Recently, increasing evidence shows that lncRNAs are involved in cellular biological processes, such as cell differentiation, proliferation, and apoptosis.

LncRNAs play important roles in the development of cancers and participate in various basic biological processes[15]. For example, lncRNA HOTAIR is highly expressed in gallbladder carcinoma tissues, causing tumor metastasis[16]. In esophageal squamous cell carcinoma, lncRNA POU3F3 is up-regulated to promote the occurrence of tumor[17,18]. LncRNA CCAL can activate the Wnt/β-catenin pathway to facilitate the progression of colorectal cancer[19,20]. Several researches have found that among malignant tumors in the blood system, AML has highly expressed LINC00319 which can be used to predicate poor prognosis and LINC00319 is confirmed to be up-regulated and exert a carcinogenic effect in multiple cancers. Meanwhile, its down-regulation inhibits the proliferation of AML cells to induce

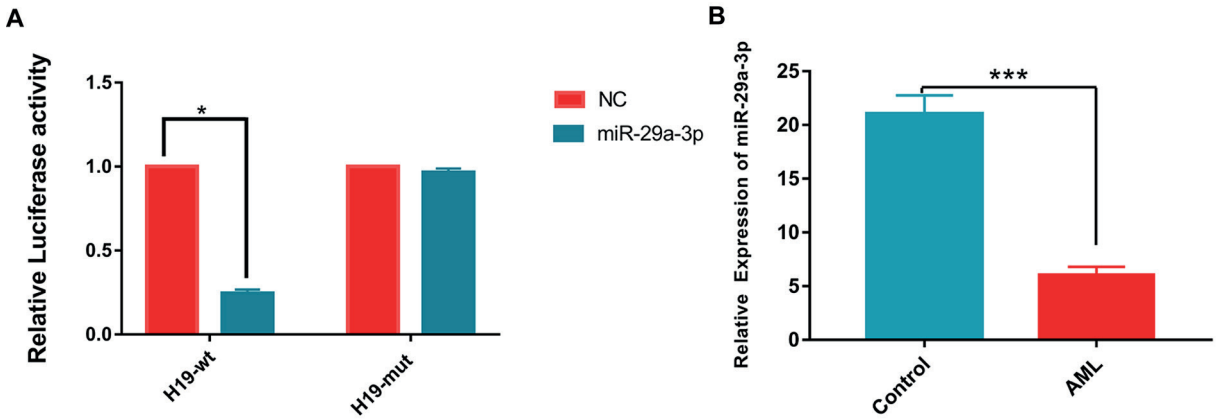

Figure 5. ***A,*** After lncRNA-H19 interacts with miR-29a-3p, the fluorescence intensity is evidently lowered ($p<0.05$). ***B,*** Expression level of miR-29a-3p in AML patients is notably lower than that in normal controls ($p<0.001$).

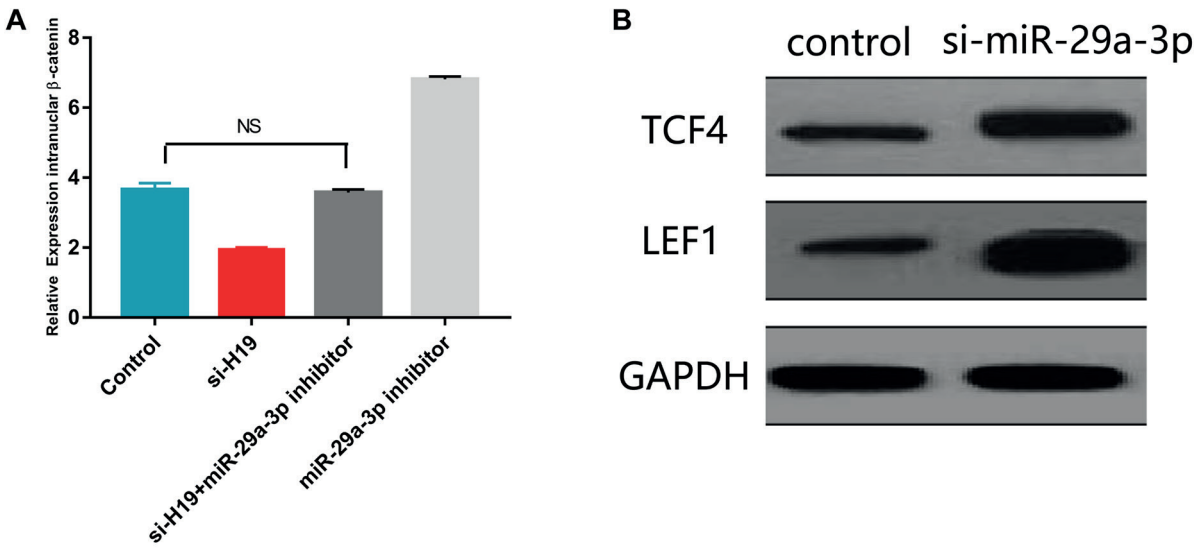

Figure 6. ***A,*** Results of qPCR show that the expression of β-catenin in the nuclei is significantly increased after the expression of miR-29a-3p is inhibited. ***B,*** According to the WB analysis, after the down-regulation of miR-29a-3p, the Wnt/β-catenin pathway critical molecules TCF and LEF1 are evidently up-regulated.

the apoptosis, thereby controlling the growth of such cells[21]. According to the findings of the recent studies, lncRNA-H19 functions in two opposite manners based on the type of tumors, namely, lncRNA-H19 functions as an oncogene in breast cancer, colorectal cancer, glioblastoma, and ovarian cancer, and as a cancer suppressive factor in liver cancer, nephroblastoma, and prostate cancer. In the present work, it was found that lncRNA-H19 functioned as an oncogene in AML, and that the up-regulation of lncRNA-H19 promoted the migration and invasion of AML cells. As described above, studies have reported the different roles of lncRNA-H19 in different types of tumors, and they are even contradictory, which means that the regulatory effect of lncRNA-H19 may be related to tumor environment. Based on this finding, it was further revealed that lncRNA-H19/miR-29a-3p/Wnt may be a potential ceRNA regulation network in AML.

MiRNAs can exert a strong regulatory function within a wide range and their important correlations with tumors have been verified in numerous studies. In 2007 AML and miRNAs were analyzed for the first time. Garaulet et al[22] found 5 kinds of specific miRNAs that can regulate the homeobox (Hox) genes through literature review based on the preliminary research on Hox genes. Havelange et al[23] discovered in subsequent studies that miR-10a is abnormally highly expressed once AML patients are accompanied by the mutation of nucleoprotamine 1. The down-regulation of miR-10a can promote the apoptosis of AML cells. According to the analysis of the dual-luciferase reporter assay, researchers have found that miR-10a suppresses two down-stream target genes, KLF4 and RB1CC1, to resist cell apoptosis. With the progress in detection techniques, the aberrantly expressed miRNAs in AML have been constantly discovered. MiRNAs will exert new effects in the diagnosis for the occurrence and development of AML, which provides a new idea for the diagnosis and targeted treatment of AML. They regulate target genes to exert their biological functions. Therefore, exploring the downstream target genes of miRNAs is the core link in the miRNA research. In this study, the bioinformatic analysis results manifested that miR-29a-3p served as the downstream target of lncRNA-H19. The inhibitory effect of miR-29a-3p on tumors has been confirmed. It was found that miR-29a-3p is down-regulated in gastric cancer, 19 cases of transparent cell renal carcinoma, 27 cases of hepatocellular carcinoma, and 28 cases of chronic myeloid leukemia[24]. Additionally, the restoration of miR-29a-3p level in PTC cell line can significantly repress tumor progression features, such as tumor cell growth, proliferation, migration, and invasion[25]. Consistent with the existing studies, the present work also found the down-regulation of miR-29a-3p in AML, which confirms its inhibitory effect on the growth and

proliferation of AML cells. These results provide another evidence to support the inhibitory effect of miR-29a-3p in cancers.

Conclusions

We revealed that lncRNA-H19 was up-regulated in AML, which is associated with the poor prognosis of AML patients. AML may promote the proliferation of AML cells by silencing the expression of miR-29a-3p. This research provides a new perspective that lncRNA-H19 plays a role as a non-coding oncogene in the occurrence of AML. Therefore, lncRNA-H19 is a new marker for early diagnosis of AML and a new target for its treatment.

Conflict of Interests

The Authors declare that they have no conflict of interests.

References

1) Saletta F, Wadham C, Ziegler DS, Marshall GM, Haber M, McCowage G, Norris MD, Byrne JA. Molecular profiling of childhood cancer: biomarkers and novel therapies. BBA Clin 2014; 1: 59-77.
2) Cummins KD, Gill S. Anti-CD123 chimeric antigen receptor T-cells (CART): an evolving treatment strategy for hematological malignancies, and a potential ace-in-the-hole against antigen-negative relapse. Leuk Lymphoma 2018; 59: 1539-1553.
3) Chen L, Mao H, Zhang J, Chu J, Devine S, Caligiuri MA, Yu J. Targeting FLT3 by chimeric antigen receptor T cells for the treatment of acute myeloid leukemia. Leukemia 2017; 31: 1830-1834.
4) Faulk K, Gore L, Cooper T. Overview of therapy and strategies for optimizing outcomes in de novo pediatric acute myeloid leukemia. Paediatr Drugs 2014; 16: 213-227.
5) Cooper TM, Franklin J, Gerbing RB, Alonzo TA, Hurwitz C, Raimondi SC, Hirsch B, Smith FO, Mathew P, Arceci RJ, Feusner J, Iannone R, Lavey RS, Meshinchi S, Gamis A. AAML03P1, a pilot study of the safety of gemtuzumab ozogamicin in combination with chemotherapy for newly diagnosed childhood acute myeloid leukemia: a report from the Children's Oncology Group. Cancer 2012; 118: 761-769.
6) Rubnitz JE, Inaba H, Dahl G, Ribeiro RC, Bowman WP, Taub J, Pounds S, Razzouk BI, Lacayo NJ, Cao X, Meshinchi S, Degar B, Airewele G, Raimondi SC, Onciu M, Coustan-Smith E, Downing JR, Leung W, Pui CH, Campana D. Minimal residual disease-directed therapy for childhood acute myeloid leukaemia: results of the AML02 multicentre trial. Lancet Oncol 2010; 11: 543-552.
7) Abrahamsson J, Forestier E, Heldrup J, Jahnukainen K, Jonsson OG, Lausen B, Palle J, Zeller B, Hasle H. Response-guided induction therapy in pediatric acute myeloid leukemia with excellent remission rate. J Clin Oncol 2011; 29: 310-315.
8) Basak P, Chatterjee S, Bhat V, Su A, Jin H, Lee-Wing V, Liu Q, Hu P, Murphy LC, Raouf A. Long non-coding RNA H19 acts as an estrogen receptor modulator that is required for endocrine therapy resistance in ER+ breast cancer cells. Cell Physiol Biochem 2018; 51: 1518-1532.
9) Liu X, Liu C, Shan K, Zhang S, Lu Y, Yan B, Luo Y. Long non-coding RNA H19 regulates human lens epithelial cells function. Cell Physiol Biochem 2018; 50: 246-260.
10) Luan W, Zhou Z, Ni X, Xia Y, Wang J, Yan Y, Xu B. Long non-coding RNA H19 promotes glucose metabolism and cell growth in malignant melanoma via miR-106a-5p/E2F3 axis. J Cancer Res Clin Oncol 2018; 144: 531-542.
11) Gong LC, Xu HM, Guo GL, Zhang T, Shi JW, Chang C. Long non-coding RNA H19 protects H9c2 cells against hypoxia-induced injury by targeting microRNA-139. Cell Physiol Biochem 2017; 44: 857-869.
12) Zhang A, Shang W, Nie Q, Li T, Li S. Long non-coding RNA H19 suppresses retinoblastoma progression via counteracting miR-17-92 cluster. J Cell Biochem 2018; 119: 3497-3509.
13) Shukla GC, Singh J, Barik S. MicroRNAs: processing, maturation, target recognition and regulatory functions. Mol Cell Pharmacol 2011; 3: 83-92.
14) Liu XH, Sun M, Nie FQ, Ge YB, Zhang EB, Yin DD, Kong R, Xia R, Lu KH, Li JH, De W, Wang KM, Wang ZX. Lnc RNA HOTAIR functions as a competing endogenous RNA to regulate HER2 expression by sponging miR-331-3p in gastric cancer. Mol Cancer 2014; 13: 92.
15) Cui H, Onyango P, Brandenburg S, Wu Y, Hsieh CL, Feinberg AP. Loss of imprinting in colorectal cancer linked to hypomethylation of H19 and IGF2. Cancer Res 2002; 62: 6442-6446.
16) Guan GF, Zhang DJ, Wen LJ, Xin D, Liu Y, Yu DJ, Su K, Zhu L, Guo YY, Wang K. Overexpression of lncRNA H19/miR-675 promotes tumorigenesis in head and neck squamous cell carcinoma. Int J Med Sci 2016; 13: 914-922.
17) Fan M, Xu J, Xiao Q, Chen F, Han X. Long non-coding RNA TCF7 contributes to the growth and migration of airway smooth muscle cells in asthma through targeting TIMMDC1/Akt axis. Biochem Biophys Res Commun 2019; 508: 749-755.
18) Hu W, Wang Z, Li Q, Wang J, Li L, Jiang G. Upregulation of lincRNA-p21 in thoracic aortic aneurysms is involved in the regulation of proliferation and apoptosis of vascular smooth muscle cells by activating TGF-beta1 signaling pathway. J Cell Biochem 2019; 120: 4113-4120.
19) Cesana M, Cacchiarelli D, Legnini I, Santini T, Sthandier O, Chinappi M, Tramontano A, Bozzoni I. A long noncoding RNA controls muscle differentiation by functioning as a competing endogenous RNA. Cell 2011; 147: 358-369.
20) Martinet C, Monnier P, Louault Y, Benard M, Gabory A, Dandolo L. H19 controls reactivation of the im-

printed gene network during muscle regeneration. Development 2016; 143: 962-971.
21) Luo W, Li G, Yi Z, Nie Q, Zhang X. E2F1-miR-20a-5p/20b-5p auto-regulatory feedback loop involved in myoblast proliferation and differentiation. Sci Rep 2016; 6: 27904.
22) Garaulet DL, Lai EC. Hox miRNA regulation within the Drosophila Bithorax complex: patterning behavior. Mech Dev 2015; 138 Pt 2: 151-159.
23) Havelange V, Ranganathan P, Geyer S, Nicolet D, Huang X, Yu X, Volinia S, Kornblau SM, Andreeff M, Croce CM, Marcucci G, Bloomfield CD, Garzon R. Implications of the miR-10 family in chemotherapy response of NPM1-mutated AML. Blood 2014; 123: 2412-2415.
24) Salati S, Salvestrini V, Carretta C, Genovese E, Rontauroli S, Zini R, Rossi C, Ruberti S, Bianchi E, Barbieri G, Curti A, Castagnetti F, Gugliotta G, Rosti G, Bergamaschi M, Tafuri A, Tagliafico E, Lemoli R, Manfredini R. Deregulated expression of miR-29a-3p, miR-494-3p and miR-660-5p affects sensitivity to tyrosine kinase inhibitors in CML leukemic stem cells. Oncotarget 2017; 8: 49451-49469.
25) Zhao Z, Wang L, Song W, Cui H, Chen G, Qiao F, Hu J, Zhou R, Fan H. Reduced miR-29a-3p expression is linked to the cell proliferation and cell migration in gastric cancer. World J Surg Oncol 2015; 13: 101.