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RESEARCH ARTICLE

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Expression of microRNAs and IRAK1 pathway genes are altered in gastric cancer patients with *Helicobacter pylori* infection

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Abstract

Gastric cancer (GC) is among the most common cancer types in the world and one of the most lethal gastrointestinal cancers. MicroRNAs (miRNAs) can be of great importance in the early detection of GC. This study aimed to investigate some miRNAs and the genes involved in IRAK1 pathways in the serum of GC patients with *Helicobacter pylori (H. pylori)* infections compared to the control group. Total RNA was extracted from the serum of GC patients with *H. pylori* infection and healthy volunteers. The expression levels of miRNAs and the genes were assessed using Real time RT-PCR with specific primers. Our data showed that miR-146, miR-375, and Let-7 were down-regulated and miR-19 and miR-21 were up-regulated in GC patients with *H. pylori* infection. Other genes involved in the pathways such as *RAS*, *MYC*, *NFKB*, *JUN*, *TRAF6,* and *IRAK4* were overexpressed; while the expression of *PTEN* gene was decreased compared to the control group. Expression of miRNAs and IRAK1 pathway genes are altered in patients with GC and *H. pylori* infection. This suggests a potential role for the above-mentioned miRNAs and genes in the diagnosis of GC.

KEYWORDS
microRNA, gastric cancer, *Helicobacter pylori*, RT-PCR

1 | INTRODUCTION

Gastric cancer (GC) is among the most common malignancies worldwide.[1,2] The burden of GC is high with approximately 14 million new cases and 8.2 million cancer-related deaths in 2012 is the prevalence of GC has been predicted to rise over the next two decades according to the World Health Organization report.[3] The most important causes of GC are *Helicobacter pylori* infection, gastric ulcer, and gastritis, smoking, and alcohol. The current gold standard of diagnosis for this cancer is histological examination of tissue obtained by a surgical excision or radiologically-guided biopsy. However, these methods are invasive, expensive, and may impose too much risk on the patients.[3] Progress in the timely

Abbreviations: GC, gastric cancer; *H. pylori*, *Helicobacter pylori*; miRNA, MicroRNA; IRAK, IL-1 receptor-associated kinases; qRT-PCR, quantitative reverse-transcriptase polymerase-chain-reaction.

diagnosis and treatment of GC has a crucial impact on the optimal management and long-term survival of the patients. Therefore, there is a need to find novel diagnostic biomarkers to allow early detection of this malignancy.[4,5] However, the molecular pathogenesis of GC still remains to be further explored. Mechanisms of tumorigenesis and progression in this disease have not yet been clarified and efficient diagnostic and prognostic biomarkers are lacking.[6,7] MicroRNAs (miRNA) are small (22 bp) noncoding regulatory nucleic acids which bind to the 3́ untranslated regions of their respective mRNAs, resulting in translational inhibition and mRNA degradation.[8] MicroRNAs are also involved in several biological functions of tumor cells including migration, proliferation, invasion, and differentiation.[9] There is evidence suggesting a connection between altered miRNA expression and cancer. Among all miRNAs that are related to cancer, let-7 is one of the earliest ones identified, and therefore has attracted most of the interest owing to its aberrant expression in human cancers.[10–12]

Recently, many studies have shown that miRNAs, which are involved in the tumorigenesis and the development of various cancers, can be detectable in the circulation.[13–15] The potential of using circulating miRNAs as a non-invasive biomarker for the detection of prostate cancer has also been reported. These reports have suggested a new and interesting field in the screening and monitoring of the cancer

IRAK signaling pathways have been reported to be related to GC. IL-1 receptor-associated kinases (IRAK) pathway consists of four family members, IRAK-1, IRAK-2, IRAK-3 (also known as IRAK-M), and IRAK-4.[16] This pathway regulates the expression of inflammatory molecules that control tumor microenvironment and promote metastasis, tumor growth, immune suppression, and chemotherapy resistance. Also, IRAK inhibitors particularly IRAK-1 and IRAK-4 have therapeutic applications in cancer. On the other hand, Wnt/β-catenin and NF-κB signaling pathways affect tumor initiation, cell proliferation, survival, tumor growth, metastasis, and resistance to therapy in GC.[17,18] MYC and RAS are among the most important oncogenes and play a key role in the regulation of cell growth and proliferation, metabolism, differentiation, apoptosis, and angiogenesis.[19] In this study, we investigated the expression of miRNAs let-7, miR-375, miR-146, miR-21 and miR-19 and several genes in IRAK1 pathway in the serum of GC patients with *H. pylori* infection, and compared these expression levels with those of healthy subjects to assess the diagnostic value of these miRNAs in GC.

2 | MATERIAL AND METHODS

2.1 | Population

In 2016-2017, pre-operative serum samples were collected from 120 GC patients with *H. pylori* infection as well as 102 healthy volunteers for the test-scale analysis. Informed consents were obtained from both groups (GC and control groups). Study approval was obtained from the Baqiyatallah University of Medical Sciences, Tehran, Iran. Three milliliter peripheral blood samples were collected from patients and healthy volunteers after obtaining an informed consent and agreement. Immediately after collection, the serum was isolated and then stored at −80°C until any further processing.

2.2 | DNA extraction for the detection *Helicobacter pylori*

Genomic double stranded DNA was extracted from peripheral blood leukocytes using QIAamp DNA Mini-Kit (Qiagen, San Diego, CA) following lysis of the cells and subsequent DNA precipitation. *H. pylori* PCR detection kit (CinnaGen, Iran) was used for the detection of *H. pylori* DNA in human serum according to the manufacturer's protocol.

2.3 | RNA extraction and cDNA synthesis

Total RNA was extracted from serum using miRNeasy serum/plasma kit, (Qiagen) according to the manufacturer's protocol. After extraction, the optical density was measured (NanoDrop 2000, Germany).

The sequence of each gene was taken from the NCBI website and was used for designing the primers listed in Table 1. miRNA sequences were taken from miRBase website and the specific stem loop primers were performed for each individual miRNA, using AlleleID software version 6 (Table 2). The sequences of primers are listed in Tables 1 and 2. Synthesis of cDNA was performed using the RevertAid First Strand cDNA Synthesis Kit Enzyme (Fermentas, Hanover, MD) according to the manufacturer's instruction.

2.4 | Quantitative reverse-transcriptase polymerase-chain-reaction (qRT-PCR)

The expression of all genes and miRNAs was performed using Syber Green qRT-PCR and specific primers. Master mix was purchased from Macrogene Company (Macrogene co, Seoul, Korea) and PCR was run using the LightCycler® 96 System Real time thermal cycler (Roche Molecular Systems, Basel, Switzerland).

Briefly, The Real time PCR reaction mixture contained 3 μL of cDNA sample, 10 μL of master mix and 1 μL of each primer (10 pmol) in a final volume of 20 mL. Quantitative PCR was carried out using the following protocol: after an initial denaturation step at 94°C for 2 min, there were 25 cycles (94°C for 15 s, 60°C for 30 s). Each sample was run and U6 (as a housekeeping gene) miRNA was used as control to normalize the expression levels of miRNAs. The mean expression values of each miRNA relative to U6 RNA were calculated using the $2^{-\Delta\Delta CT}$ method (16).

TABLE 1 The sequence of primers for genes in the IRAK1 pathway

Gen	Primer	Sequence (5′-3′)
GAPDH	FP	5′-CCACTCCTCCACCTTTGAC-3′
	RP	5′-ACCCTGTTGCTGTAGCCA-3′
RAS	FP	5′-CGCCTCGCAAGACTCCAG-3′
	RP	5′-CAGCAGAAGGTGATCCAGACT-3′
MYC	FP	5′-CGCCTCGCAAGACTCCAG-3′
	RP	5′-CAGCAGAAGGGAATCCAGACTC-3′
NFKB	FP	5′-GCTGAGTCCTGCTACTTCC-3′
	RP	5′-CGTCTATTTGCTCGATTGTGG-3′
JUN	FP	5′-GCGGACCCTATGGCTACAGTAAC-3′
	RP	5′-GTCGGCGTGGTGGTGATG-3;
TRAF6	FP	5′-TCTGAAAGTGACTGCTGTGTG-3′
	RP	5′-ACTGCTTCTCTGAATGCCTAC-3′
IRAK-4	FP	5′-TTGAGCAGATGTCAGTCATGGA-3′
	RP	5′-TTACCGCTCCGAGCTTCTTC-3′
PTEN	FP	5′-CCAATGGCTAAGTGAAGATGAC-3′
	RP	5′-TCCAGATGATTCTTTAACAGGTAG-3′

(FP), Forward primer; (RP), Reverse primer.

Using the standard curves drawn for each primer, the generated fluorescence intensity was recorded and the corresponding calculations were performed. For each sample, the expression level of miRNA U6 was also examined for normalization. To compare the patient samples with the healthy control samples, the expression of each gene was expressed as the ratio of expression to the normal U6 miRNA gene, and the final relative expression was expressed using

TABLE 2 Sequence of MicroRNA primers

miRNA	Primer stem loop	Sequence (5′-3′)
miR-21	RT	5′-GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACTCAACA-3′
	FP	5′-GCCGCTAGCTTATCAGACTGATGT-3′
	RP	5′-GTGCAGGGTCCGAGGT-3′
U6	RT	5′-GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACAAAAATATG-3′
	FP	5′-GCGCGTCGTGAAGCGTTC-3′
	RP	5′-GTGCAGGGTCCGAGGT-3′
miR-146a	RT	5′-GTCGTATTGCAGAGCAGGGTCAGAGGTATTCGCAATGCAGACAACCCAT-′3
	FP	5′-TCCGTGAGAACTCAATTCC-3′
	RP	5′-GAGCAGGGTCCGAGGT-3′
miR-375	RT	5'-GTCGTATGCAGAGCAGGGTCGGAGGTATTCGCACTGCATACGACTCACGC-'3
	FP	5′-ACTTTGTTCGTTCGGCTC-′3
	RP	5′-GAGCAGGGTCGGAGGT-′3
miR-19	RT	5′-GTCGTATGCAGAGCAGGGTCCGAGGTATTCGCACTGATCACGACTCGATT-′3
	FP	5′-CGTGTGACAGGTTGACCA-′3
	RP	5′-CGGTGAGCAAATGTATGCAA-′3
miR-LET-7a	RT	5′-GAGCAGGGTCCGAGGTTCGCACTGCATACGACTCACGC-'3
	FP	5′-GAGCGGATTCAGATAACCAAGC-′3
	RP	5′-CCATCCAGTGTACTTGCTACAG-′3

TABLE 3 comparison of the baseline characteristics between control or case group

	Control (*N*)	Case (*N*)	*P*-value
Age			
Male	42.5 ± 3.29	66.52 ± 15.62	0.001
Female	51.24 ± 7.23	61.56 ± 17.64	
Frequency *N*(%)			
Male	52 (51%)	79 (65.8%)	0.03
Female	50 (49%)	41 (34.2%)	
Total	102 (100%)	120 (100%)	

Values are expressed as mean ± SD, median.

the ΔΔCT formula. The expression of all genes in the messenger pathway was also normalized by the GAPDH gene.

2.5 | Statistical analysis

Statistical analyses were performed using the SPSS software version 16 for Windows (SPSS Inc., Chicago, IL). The $P \leq 0.05$ was considered as statistically significant.

3 | RESULTS

A total of 222 subjects, including 120 GC patients and 102 healthy controls, were recruited in this study. Age and gender distribution of the study groups are shown in Table 3.

The healthy control group consisted of a total of 102 subjects (50 females and 52 males) with a mean age of 51.24 ± 7.23 (females) and 42.5 ± 3.29 (males) years. The GC patients group were all infected with *H. pylori* and included 120 subjects (41 females and 79 males) with a mean age of 61.56 ± 17.64 (females) and 66.52 ± 15.62 (males) years. The age distribution of the patient group is shown in Table 4.

Detection of *H. pylori* infection in GC patients based on gel electrophoresis is shown in Figure 1.

TABLE 4 Age distribution of the patients group with gastric cancer and *H. pylori* infection

Age groups (year)	Total percentage	Percentage in male/female
35-44	9.16	36.36/63.63
45-54	16.68	65/35
55-64	19.16	69.56/30.43
65-74	25	63.33/36.66
75-84	20	75/25
85≤	10	75/25

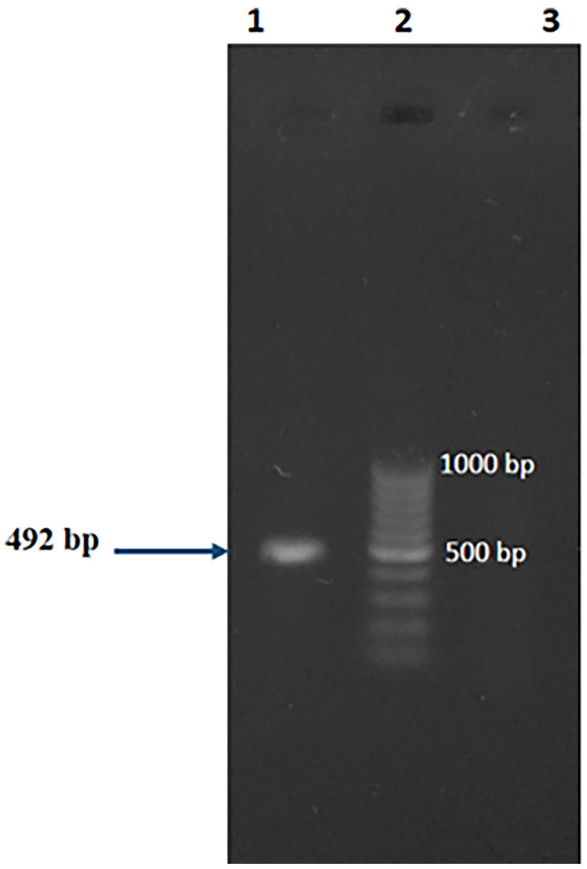

FIGURE 1 Gel electrophoresis of *Helicobacter pylori* detection. Lane 1, *H. pylori* positive; lane 2, 100 bp DNA size-marker (Fermentas); lane 3, *H. pylori* negative

Expression of miRNAs in the serum of GC patients with *H. pylori* infection and healthy controls were compared (Figures 2 and 3). We analyzed five miRNAs including let-7, miR-375, miR-146, miR-21, and miR-19. Among the five miRNAs analyzed, the expression of miRNAs 19 and 21 were up-regulated in patients compared with controls (Figure 2) while miRNAs 375, 146, and let-7 were down-regulated in patients (Figure 3).

We also examined the expression of *RAS*, *MYC*, *NF-κB*, *JUN*, *TRAF6,* and *IRAK4* which showed overexpression. On the other hand, *PTEN* expression was found to be reduced. These genes are involved in the expression of miRNAs 19, 21, 375, 146, and let-7 (Figure 4).

4 | DISCUSSION

Numerous genetic alterations have been identified to be involved in the tumorigenesis of various cancer types. Recent studies have shown the potential of circulating nucleic acids as non-invasive biomarkers of different cancers.[20,21] During the past years, miRNAs have been suggested to serve as potential biomarkers for

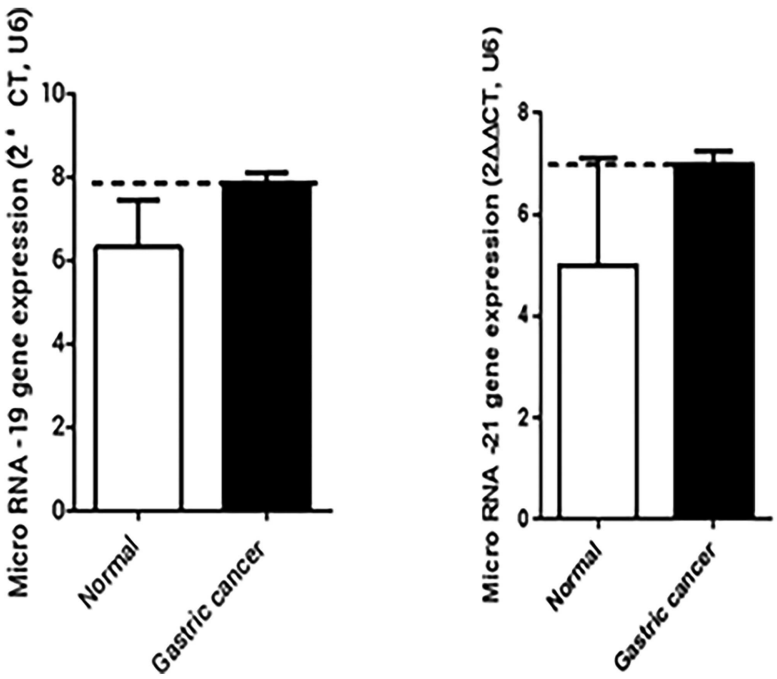

FIGURE 2 Up-regulation of miR-19 and miR-21 in patients compared to control group. *$P < 0.05$, compared with the control group

tumor progression and oncogenesis.[21–23] MiRNAs can be detected in various body fluids such as serum, plasma, and urine. This property of circulating miRNAs can introduce them as potential biomarkers of cancers.[24] In the present study, we assessed five miRNAs including miR-19, miR-21, miR-375, miR-146, and Let-7 in GC patients with *H. pylori* infection. In our study, miR-19 and miR-21 were up-regulated while miR-375, miR-146, and let-7 were down-regulated. Let-7 has been suggested to serve as a tumor repressor and to be strongly associated with clinic-pathological factors and prognosis of cancers.[10–12] Let-7 is involved in several pathological processes such as apoptosis, proliferation, and invasion of cancer cells. The signaling pathways and oncogenes are a potential connection between the level of expression of let-7 and the biological feature of tumor cells.[24] Several previous studies have reported that let-7 is down regulated in GC.[12,25–27] These studies, along with the

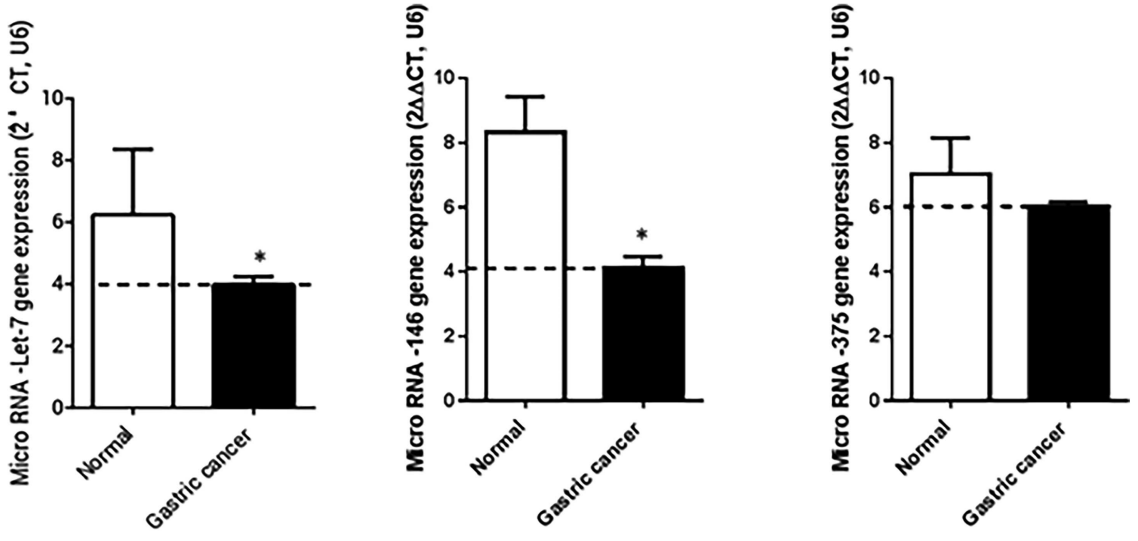

FIGURE 3 Down-regulation of microRNAs Let-7, miR-375, and miR-146 in patients compared to control group. *$P < 0.05$, compared with the control group

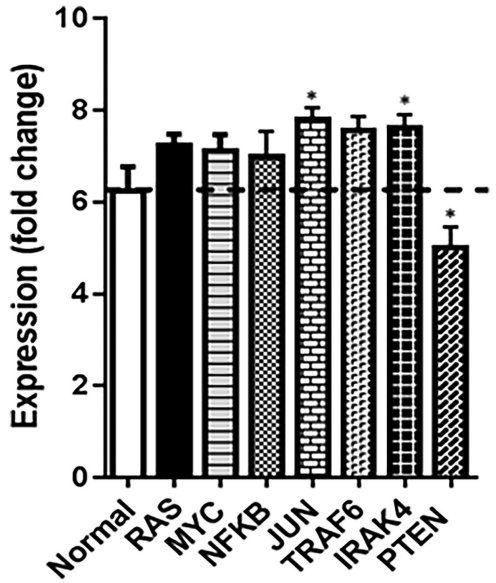

FIGURE 4 Expression of IRAK signaling pathway in patients with gastric cancer and *H. pylori* infection compared to control group, $*P < 0.05$: compared with the control group

present results, suggested that the reduced let-7 expression is a frequent event in GC. MiR-375 is down regulated in many types of cancer and inhibits cancer progression by targeting several key oncogenes. In our study, miR-375 was down regulated which is similar to the finding of previous studies.[28] that the role of miR-375 as a tumor suppressor in GC has also been reported.[29] In another study, miR-375 was found to be down-regulated in response to *H. pylori* infection. *H. pylori* infection is the most prevalent infection in the world and has been identified as the main cause of GC. A subset of miRNAs has been reported to be associated with *H. pylori* infection, as among which are miR-141, miR-146, and miR-375.[30] In the present study, we examined miRNAs expression in the serum of the patients with GC and *H. pylori* infections. Our results indicated that miR-146 was down-regulated in GC, which is consistent with the findings of previous studies.[7] *H. pylori* infection is associated with some miRNAs, one of them is miR-146 which is reported in our study.[31] It has been reported that miR-146 functions as a tumor suppressor in GC and it inhibits tumor progression by targeting EGFRs, CXCR4, and IRAK1.[31–33] We found that IRAK1 is overexpressed, a gene that is inhibited by miR-146 according to a previous study.[31] Ryunosuke et al, indicated that the reduced expression of miR-146 plays a role in GC progression through lymph node metastasis by the inhibition of EGFR and IRAK1.[31] MiR-21 acts as an oncogene because most of its target genes are tumor suppressors. Up regulation of miR-21 in different cancers, and its function in targeting a variety of tumor suppressors genes suggest the use of this miRNA as a diagnostic and prognostic biomarker.[34] In the present study, miR-21 was found to be up-regulated, which is consistent with previous studies.[3,21,35] The negative relationship between miR-21 and its targets PTEN and PDCD4 is already established.[36] Our results confirmed that up regulation of miR-21 reduces the expression of *PTEN* in GC patients with *H. pylori* infection. Recently, a study showed that miR-19 is up regulated in GC.[5] The function of miR-19 in GC has not yet been fully defined but it has been suggested that miR-19 up regulation might play a significant role in the progression of GC as well as cell proliferation and colony formation.[37] In our study, miR-19 was up regulated which is similar to previous studies. MYC is the most studied oncogene owing to its association with several diseases.[38] MYC plays a significant role in the regulation of cell growth, proliferation, differentiation, and apoptosis.[39] MYC amplification seems to be associated with GC.[38] Another study reported that c-MYC overexpression is found in over 40% of GC patients.[40] In the present study, *MYC* gene was overexpressed in GC patients and our results were similar to the previous studies.

5 | CONCLUSION

This study indicated that miR-146, miR-375, and let-7 were down-regulated while miR-19 and miR-21 were up-regulated in GC patients with *H. pylori* infection. IRAK signaling pathway, Wnt/β-catenin and NF-κB signaling were also found to be deregulated in GC patients with *H. pylori* infection.

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CONFLICTS OF INTEREST

The authors have no conflict of interest to disclose.

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REFERENCES

1. Ferlay J, Shin HR, Bray F, Forman D, Mathers C, Parkin DM. Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. *Int J Cancer*. 2010;127:2893–2917.
2. Yepes S, López R, Andrade RE, Rodriguez-Urrego PA, López-Kleine L, Torres MM. Co-expressed miRNAs in gastric adenocarcinoma. *Genomics*. 2016;108:93–101.
3. Larrea E, Sole C, Manterola L, et al. New concepts in cancer biomarkers: circulating miRNAs in liquid biopsies. *Int J Mol Sci*. 2016;17:627.
4. Bloomston M, Frankel WL, Petrocca F, et al. MicroRNA expression patterns to differentiate pancreatic adenocarcinoma from normal pancreas and chronic pancreatitis. *JAMA*. 2007;297:1901–1908.

5. Ueda T, Volinia S, Okumura H, et al. Relation between microRNA expression and progression and prognosis of gastric cancer: a microRNA expression analysis. *Lancet Oncol*. 2010;11:136–146.
6. Li X, Luo F, Li Q, et al. Identification of new aberrantly expressed miRNAs in intestinal-type gastric cancer and its clinical significance. *Oncol Rep*. 2011;26:1431–1439.
7. Liu D, Hu X, Zhou H, Shi G, Wu J. Identification of aberrantly expressed miRNAs in gastric cancer. *Gastroenterol Res Pract*. 2014;2014.
8. Nana-Sinkam S, Croce C. Clinical applications for microRNAs in cancer. *Clin Pharmacol Ther*. 2013;93:98–104.
9. Shimono Y, Zabala M, Cho RW, et al. Downregulation of miRNA-200c links breast cancer stem cells with normal stem cells. *Cell*. 2009;138:592–603.
10. Takamizawa J, Konishi H, Yanagisawa K, et al. Reduced expression of the let-7 microRNAs in human lung cancers in association with shortened postoperative survival. *Cancer Res*. 2004;64:3753–3756.
11. Akao Y, Nakagawa Y, Naoe T. Let-7 microRNA functions as a potential growth suppressor in human colon cancer cells. *Biol Pharm Bull*. 2006;29:903–906.
12. Yang Q, Jie Z, Cao H, et al. Low-level expression of let-7a in gastric cancer and its involvement in tumorigenesis by targeting RAB40C. *Carcinogenesis*. 2011;32:713–722.
13. Calin GA, Croce CM. MicroRNA signatures in human cancers. *Nat Rev Cancer*. 2006;6:857–866.
14. Filipowicz W, Bhattacharyya SN, Sonenberg N. Mechanisms of post-transcriptional regulation by microRNAs: are the answers in sight? *Nat Rev Genet*. 2008;9:102–114.
15. Thammaiah CK, Jayaram S. Role of let-7 family microRNA in breast cancer. *Non-coding RNA Res*. 2016;1:77–82.
16. Castaño-Rodríguez N, Kaakoush NO, Pardo AL, Goh K-L, Fock KM, Mitchell HM. Genetic polymorphisms in the Toll-like receptor signalling pathway in *Helicobacter pylori* infection and related gastric cancer. *Hum Immunol*. 2014;75:808–815.
17. Hsieh Y-Y, Shen C-H, Huang W-S, et al. Resistin-induced stromal cell-derived factor-1 expression through Toll-like receptor 4 and activation of p38 MAPK/NFκB signaling pathway in gastric cancer cells. *Int J Biomed Sci*. 2014;21:59.
18. Chiurillo MA. Role of the Wnt/β-catenin pathway in gastric cancer: An in-depth literature review. *World J Exp Med*. 2015;5:84.
19. Li T, Mo X, Fu L, Xiao B, Guo J. Molecular mechanisms of long noncoding RNAs on gastric cancer. *Oncotarget*. 2016;7:8601.
20. Sidransky D. Nucleic acid-based methods for the detection of cancer. *Science*. 1997;278:1054–1058.
21. Tsujiura M, Ichikawa D, Komatsu S, et al. Circulating microRNAs in plasma of patients with gastric cancers. *Br J Cancer*. 2010;102:1174–1179.
22. He L, Thomson JM, Hemann MT, et al. A microRNA polycistron as a potential human oncogene. *Nature*. 2005;435:828–833.
23. Lu J, Getz G, Miska EA, et al. MicroRNA expression profiles classify human cancers. *Nature*. 2005;435:834–838.
24. Mitchell PS, Parkin RK, Kroh EM, et al. Circulating microRNAs as stable blood-based markers for cancer detection. *Proc Natl Acad Sci USA*. 2008;105:10513–10518.
25. Wang X, Cao L, Wang Y, Wang X, Liu N, You Y. Regulation of let-7 and its target oncogenes. *Oncol Lett*. 2012;3:955–960.
26. Tsai M-M, Wang C-S, Tsai C-Y, et al. Potential diagnostic, prognostic and therapeutic targets of microRNAs in human gastric cancer. *Int J Mol Sci*. 2016;17:945.
27. Zhang H-H, Wang X-J, Li G-X, Yang E, Yang N-M. Detection of let-7a microRNA by real-time PCR in gastric carcinoma. *World J Gastroenterol*. 2007;13:2883.
28. Lee SW, Park KC, Kim JG, et al. Dysregulation of microRNA-196b-5p and microRNA-375 in gastric cancer. *Gastric Cancer*. 2016;16:221–229.
29. Tsukamoto Y, Nakada C, Noguchi T, et al. MicroRNA-375 is downregulated in gastric carcinomas and regulates cell survival by targeting PDK1 and 14-3-3ζ. *Cancer Res*. 2010;70: 2339–2349.
30. Miao L, Liu K, Xie M, Xing Y, Xi T. MiR-375 inhibits *Helicobacter pylori*-induced gastric carcinogenesis by blocking JAK2-STAT3 signaling. *Cancer Immunol Immunother*. 2014;63: 699–711.
31. Kogo R, Mimori K, Tanaka F, Komune S, Mori M. Clinical significance of miR-146a in gastric cancer cases. *Clin. Cancer Res*. 2011;17:4277–4284.
32. Bhaumik D, Scott GK, Schokrpur S, et al. MicroRNAs miR-146a/b negatively modulate the senescence-associated inflammatory mediators IL-6 and IL-8. *Aging*. 2009;1:402.
33. Hurst DR, Edmonds MD, Scott GK, Benz CC, Vaidya KS, Welch DR. Breast cancer metastasis suppressor 1 up-regulates miR-146, which suppresses breast cancer metastasis. *Cancer Res*. 2009;69:1279–1283.
34. Kurdistani ZK, Saberi S, Tsai K-W, Mohammadi M. MicroRNA-21: mechanisms of oncogenesis and its application in diagnosis and prognosis of gastric cancer. *Arch Iran Med*. 2015;18:524.
35. Effatpanah H, Yadegarazari R, Karami M, Majlesi A, Shabab N, Saidijam M. Expression analysis of mir-21 and mir-221 in cancerous tissues from Iranian patients with gastric cancer. *Iran Biomed J*. 2015;19:188.
36. Li L, Zhou L, Li Y, Lin S, Tomuleasa C. MicroRNA-21 stimulates gastric cancer growth and invasion by inhibiting the tumor suppressor effects of programmed cell death protein 4 and phosphatase and tensin homolog. *J Buon*. 2014;19: 228–236.
37. Qin S, Ai F, Ji W-F, Rao W, Zhang H-C, Yao W-J. MiR-19a promotes cell growth and tumorigenesis through targeting SOCS1 in gastric cancer. *Asian Pac J Cancer Prev*. 2013;14: 835–840.
38. de Souza CRT, Leal MF, Calcagno DQ, et al. MYC deregulation in gastric cancer and its clinicopathological implications. *PLoS ONE*. 2013;8:e64420.
39. Pelengaris S, Khan M. The many faces of c-MYC. *Arch Biochem Biophys*. 2003;416:129–136.
40. Calcagno DQ, Leal MF, Assumpção PP, Smith MdAC, Burbano RR. MYC and gastric adenocarcinoma carcinogenesis. *World J Gastroenterol*. 2008;14:5962.

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