BRITISH JOURNAL OF BIOMEDICAL SCIENCE
2020, VOL. 77, NO. 2, 76–80
https://doi.org/10.1080/09674845.2019.1692762

Association of rs2620381 polymorphism in miR-627 and gastric cancer

M Raad [a], Z Salehi [a], M Habibzaadeh Baalsini [a], F Mashayekhi [a] and H Saeidi Saedi [b]

[a]Department of Biology, Faculty of Sciences, University of Guilan, Rasht, Iran; [b]Department of Radiation Oncology, Cancer Research Center, Guilan University of Medical Sciences (GUMS), Rasht, Iran

ABSTRACT

Background: Gastric cancer is a complicated malignancy whose aetiology is not well characterized. Single nucleotide polymorphisms (SNPs), some located within microRNA genes, are linked with gastric cancer. We hypothesized a link between SNP rs2620381 (A > C) in miR-627 and gastric cancer.
Material and methods: We recruited 280 healthy controls and 240 gastric cancer patients. Genotyping was conducted by allele-specific polymerase chain reaction. In addition, *in silico* analyses were carried out via databases and web tools including miRBase, dbSNP, RNAfold, MiRNASNP V2.0, miRWalk V2.0, miRTarBase, and miRmap.
Results: Any C genotype in rs2620381 was linked to gastric cancer: CC vs. AA: OR/95% CI 2.67 (1.17–6.09), p = 0.01, CC+AC vs. AA: OR/95% CI 1.66 (1.12–2.46), p = 0.01, CC vs. AC+AA: OR/95% CI 2.44 (1.07–5.54), p = 0.03. The minor allele C of miR-627 was linked with gastric cancer compared with A allele (OR/95%CI 1.88 (1.30–2.73), p = 0.0008). There were no links between age, sex, tumour type, distant metastasis, and tumour stages and the miR-627 polymorphism in gastric cancer patients.
Conclusion: Presence of the C SNP in miR-627 rs2620381 in linked with gastric cancer, and may be important in pathogenesis.

ARTICLE HISTORY
Received 9 October 2019
Accepted 11 November 2019

KEYWORDS
Gastric cancer; miR-627; polymorphism; in silico analysis; SNP; microRNA

Introduction

Although the 5th highest number of new cancers worldwide, gastric cancer has the third highest rate of death [1]. *H. pylori* infection is the most important factor in the occurrence of gastric cancer, although only a small number of infected individuals develop the disease, indicating that other risk factors, including genetic susceptibility and environment, are involved [2,3]. Although gastric cancer is a complex disease and its pathogenesis mechanism is not completely understood, emerging studies have shown that genetic variations, especially polymorphisms related to microRNAs (miRNAs), are significantly linked to the risk of the disease [4].

miRNAs are small endogenous noncoding RNAs, with a length of 18–25 ribonucleotides that play important roles in cancer-related biological mechanisms including cell proliferation, apoptosis, angiogenesis, migration, and invasion [5,6], and can act as tumour suppressors or oncogenes by targeting various genes involved in carcinogenesis [7]. miRNAs interact with the 3′ untranslated region (3`UTR) of target mRNAs by their 'seed region' (miRNA nucleotides 2–7), thereby regulating gene expression at the post-transcriptional level through suppressing translation, directly cleaving target mRNA, or reducing the stability of mRNAs [8–10]. Some miRNAs, such as miR-627 and miR-18a, have associated with the gastric cancer diagnostic and susceptibility [11,12]. miR-627 transcribed from genomic loci 15q15.1, processes to two functional mature miRNAs, miR-627-5p and miR-627-3p.

Single nucleotide polymorphisms (SNPs) are the most frequent form of genetic variations in the human genome which can have an impact on cancer predisposition [13,14]. SNPs located at pre-miRNAs or regions of mature miRNAs known as miR-SNPs may influence on the interaction between miRNA and target mRNA by changing the binding affinity of miRNA to their target mRNA or pre-miRNA maturation process, and so can be effective in susceptibility to different diseases, including gastric, lung, hepatocellular, colorectal, and breast cancers [15–21]. miR-627 plays several significant roles in the development of some cancers [12,22]. rs2620381 (A > C) is a common SNPs located at pre-miR-627 and may affect its function. Since miR-627 may be related to increased risk of gastric cancer by targeting of many genes involved in cancer pathways, we hypothesized an association between the rs2620381 (A > C) SNP and gastric cancer.

Material and methods

We tested our hypothesis on 240 cases of histologically-proven gastric adenocarcinoma, collected between January 2015 and August 2017. Exclusion criteria were a prior history of cancer and chemo-or radiotherapy. In addition, 280 healthy controls who had never been diagnosed with gastric adenocarcinoma, other carcinomas, or other serious diseases were selected. Control subjects were also genetically unrelated to the case subjects. All subjects completed a short questionnaire including

CONTACT Z Salehi geneticzs@yahoo.co.uk Department of Biology, Faculty of Sciences, University of Guilan, Rasht, Iran

questions on age, family history of malignancy, and chemo-or radiotherapy. Written informed consent for the genetic investigations was obtained from each subject participating in this research. The research was carried out in agreement with the Declaration of Helsinki regarding the utilization of human samples.

Genomic DNA of each participant was extracted from peripheral blood samples using the GPP Solution kit (Gene Pajoohan Pouya, Iran). The quality and quantity of the extracted DNA were confirmed using the 1% agarose gel and the Nanodrop One Microvolume UV-Vis Spectrophotometer (Thermo Fisher Scientific, USA), respectively. Genotyping of rs2620381 polymorphism was conducted by allele-specific PCR. The fragments containing A allele and C allele were multiplied using the sequences of the primers (F: 5′-CTCCTCTTTTCTTAGAGACGCA-3′ and R: 5′-TGATTTCTGATTACAAGCCCCA-3′) and (F 5′-CTCCTCT TTTCTTAGAGACGCC-3′ and R: 5′-TGATTTCTGATTACA AGCCCCA-3′), respectively. The PCR reaction was conducted in a total volume of 25µl containing 2 µl $MgCl_2$, 1 µl PCR buffer (10×), 0.5 µl dNTPs, 0.5 µl each primer, 0.2 µl Taq DNA polymerase (Kawsar Biotech Company, Iran) and 3.5 µl genomic DNA as a template. The reaction mixtures were denatured at 95 °C for 5 min, followed by 31 cycles of denaturation at 95 °C for 45 s, annealing at 53 °C for 45 s, and elongation at 72 °C for 45 s, with a final elongation at 72 °C for 5 min for A allele. The reaction condition for C allele was the same as described for A allele (except annealing at 55 °C). The amplified products were analysed using 2% agarose gel electrophoresis. The size of the PCR products of both A and C alleles were 446 bp.

Pearson's Chi-square (χ2) test was used to assess the difference in genetic distributions of genotype and allele frequencies between patients and the controls. The significance of association was evaluated by odds ratios (OR) with 95% confidence intervals (95%CI). P-value < 0.05 was considered statistically significant. All statistical analyses were carried out using MedCalc statistical software (version 15.8).

According to the miRBase database [23], miR-627 has two mature miRNA (miR-627-3p and miR-627-5p), which miR-627-5p is the predominant product of miR-627 stem-loop. By using the dbSNP database [24] and MiRNASNP V2.0 Database [25], we find the exact location of rs2620381 polymorphism on miR-627-5p. The minor allele frequencies were obtained from dbSNP. To examine whether the rs2620381 polymorphism can affect the secondary structure of miR-627, we used RNAfold web server [26]. Moreover, we used miRWalk V2.0 [27] and miRTarBase [28] databases to determine validated targets of miR-627-5p and then related target genes to gastric cancer were extracted by searching through previous studies. miRmap web tool [29] was used to evaluate the effects of the SNP within the seed site of miR-627-5p on the miRNA-mRNA interaction.

Results

Of the 240 patients with gastric adenocarcinoma, 161 were male and 79 were female, whilst 174 and 106 of the healthy controls were male and female respectively (p = 0.3). The mean [SD] age of 240 patients and 280 controls were 54.5 ± [10.9] and 55.4 ± 11.9 years, respectively (t test p=0.27). The demographic and clinic-pathological characteristics of the cases according to rs2620381 genotypes are given in Table 1. Type of cancer was defined by Lauren's classification [30]. There was no significant relationship between age, sex, tumour type, distant metastasis, and tumour stages and rs2620381 polymorphism. The genotype frequency distribution of rs2620381 in the cases and the controls are shown indicated in Table 2. Presence of the C SNP was strongly linked to the presence of gastric cancer.

Table 1. Relationship of clinic-pathological status and rs2620381 genotypes in gastric cancer patients.

	Genotype			
	AA	AC	CC	
Feature	N(%)	N(%)	N(%)	*p*
Sex				
Female	59 (35.97)	17 (29.31)	3 (16.66)	0.2
Male	105 (64.02)	41 (70.68)	15 (83.33)	
Age				
<50	45 (27.43)	20 (34.48)	8 (44.44)	0.24
≥50	119 (72.56)	38 (65.51)	10 (55.55)	
Type[a]				
Intestinal	123 (75)	37 (63.79)	11 (61.11)	0.16
Diffuse	41 (25)	21 (36.2)	7 (38.88)	
Distant metastasis				
M0	120 (73.17)	36 (62.06)	11 (61.11)	0.2
M1	44 (26.82)	22 (37.93)	7 (38.88)	
Stage				
I	23 (14.02)	3 (5.17)	0	0.06
II	57 (34.75)	20 (34.48)	3 (16.66)	
III	40 (24.39)	13 (22.41)	8 (44.44)	
VI	44 (26.82)	22 (37.93)	7 (38.88)	

Table 2. Genotype and allele frequencies of rs2620381 polymorphism in cases and controls.

	Cases	Control		
Model/Genotype	n (%)	n (%)	OR (95% CI)	*p*
Codominant	164	219	1.00 (Ref.)	–
AA	(68.3)	(78.2)		
AC	58	52	1.48	0.06
	(24.2)	(18.6)	(0.97–2.27)	
CC	18	9	2.67	0.01
	(7.5)	(3.2)	(1.17–6.09)	
Dominant	164	219	1.00 (Ref)	–
AA	(68.3)	(78.2)		
AC+CC	76	61	1.66	0.01
	(31.7)	(21.8)	(1.12–2.46)	
Recessive	222	271	1.00 (Ref)	–
AC+AA	(92.5)	(96.8)		
CC	18	9	2.44	0.03
	(7.5)	(3.2)	(1.07–5.54)	
Overdominant	182	228	1.00 (Ref.)	–
CC+AA	(75.8)	(81.4)		
AC	58	52	1.39	0.12
	(24.2)	(18.6)	(0.91–2.13)	
Allele	386	490	1.00 (Ref)	–
A allele	(80.4)	(87.5)		
C allele	94	70	1.7	0.001
	(19.6)	(12.5)	(1.21–2.38)	

OR = odds ratio; CI = confidence interval.

In silico analysis

rs2620381 polymorphism A > C [with global minor allele frequency (Global MAF) 0.08] was located in the first nucleotide of miR-627-5p seed site, which can change U nucleotide (in the wild-type form) to G nucleotide (in the variant form) in the miRNA sequence. Validated targets of miR-627-5p as the main mature variant of miR-627 have been identified by using miRWalk and miRTarBase databases. It has been identified that miR-627-5p has 70 validated targets including *USP42, ESCO2, DUSP5, WEE1, BTF3L4, KDM3A, NUTM2E, METRN, SEPHS1, NKX6-1, HSBP1, LUZP1, OGT, CD1D, RNF165, ERP44, XPO7, ST6GAL1, FAM168A, ZNF623, CENPN, PTPRB, SHMT2, P3H2, PHF5A, FOS, CARNMT1, APH1A, KANSL1, NAV1, AGO2, C17orf105, EFNB2, HSPA1B, TMEM98, FLVCR1, SURF4, CEBPG, PLD5, RPS16, Slfn5, SLFN5, KLHL12, ZNF548, OGFRL1, ANTXR2, JMJD1C, CLIC6, PPP1R16B, CYB561A3, MSRB1, ICOSLG, CORO2A, TMX4, CDKAL1, COX6B1, GEMIN4, USP15, ATXN7L3B, TFRC, INSIG1, IPPK, ANKRD46, CBX8, RBM20, BTG2, CKAP2L, FGFR1OP, KNSTRN*, and *ST18*. To better understand the connection between mir-627-5p targets with gastric cancer, by searching through previous researches, we found that 11 of these genes (*BTG2, AGO2, USP42, ESCO2, DUSP5, CD1D, EFNB2, SLFN5, INSIG1, FOS*, and *WEE1*) were linked with gastric cancer

In silico analysis using miRmap web tool reveals that miR-627-5p binds to the target mRNAs with weaker binding affinity in the presence of the variant allele compared to the wild-type allele. In addition, in most cases, the binding of miR-627-5p is absolutely disrupted in the presence of the variant allele (Table 3). Furthermore, evaluating the influence of the variant allele on the stem-loop structure stability of miRNA by RNAfold web server indicated that the structure minimum free energy (MFE) in the presence of variant allele was −51.66 kcal/mol in compared to −56.40 kcal/mol in the presence of wild-type allele (Figure 1).

Table 3. miRmap web tool result.

	ΔG total	
Target genes	Wild-type allele (A allele)	Variant allele (C allele)
BTG2	−0.86	Lost
AGO2	5.92	Lost
USP42	0.81	Lost
ESCO2	−12.3	0.37
DUSP5	−6.55	11.85
CD1D	0.42	14.42
EFNB2	5.51	Lost
SLFN5	−0.15	0.92
INSIG1	−0.42	1.34
FOS	9.51	Lost
WEE1	0.44	Lost

More negative ΔG means stronger interaction between miRNA and target genes. Lost means the interaction between miRNA and target gene is completely disrupted.

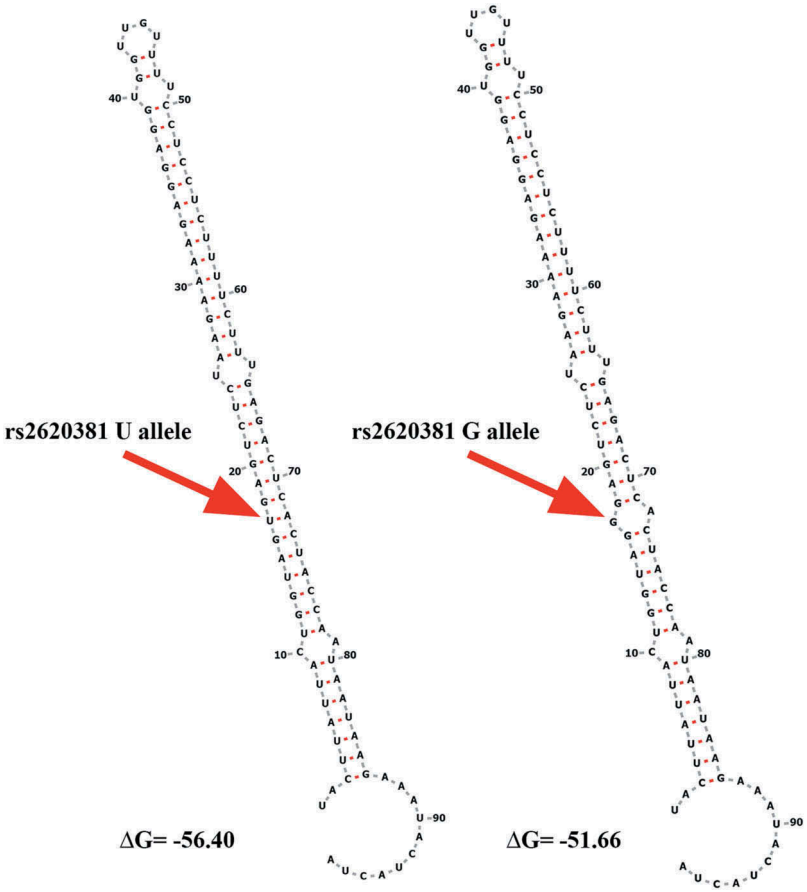

Figure 1. miR-627 wild-type (U) and variant (G) allele secondary structure prediction by RNAfold web server.

Discussion

In the present case-control study with 240 gastric cancer patients and 280 cancer-free controls, we hypothesized links between a rs2620381 SNP and this malignancy. The results of the current study revealed a significant link between miR-627 polymorphism (A > C) and gastric cancer. Moreover, the presence of rs2620381 was not linked to age, sex, tumour type, distant metastasis, and tumour stages in gastric cancer patients.

MicroRNAs play substantial roles in regulating several biological processes, such as differentiation, apoptosis, cell proliferation, tumourigenesis, inflammation, and migration [31–34]. Many studies have focused on the relation of microRNAs SNPs with gastric cancer. A number of association researches have been carried out to assess the relationship between rs2620381 SNP and the risk of various types of cancer in different populations. Li et al. observed that rs2620381 in miR-627 is not associated with the risk of lung cancer [35], whilst Zhang et al. showed that rs2620381 in miR-627 is not associated with oesophageal squamous cell carcinoma [36]. Many molecular studies have examined the function of miR-627 in different biological pathways and diseases. Sun et al. demonstrated that vitamin D increases the effect of Irinotecan via miR-627-mediated inhibition of intratumoural drug metabolism [37]. Padi et al. indicated that miR-627 mediates vitamin D tumour-suppressive activities in human colorectal cancer cells by targeting histone demethylase *JMJD1A* [22]. Furthermore, Chen et al. demonstrated that cell proliferation and epithelial-to-mesenchymal transition are regulated by LINC00958 through targeting miR-627 in oral squamous cell carcinoma [38]. Cao et al, found that miR-627 can down-regulate the expression of *BRCA2* in breast cancer [39], whilst Shin et al. reported it to be significantly up-regulated in gastric cancer [12].

miR-SNPs can affect the function of miRNA through two mechanisms. First, miR-SNPs can affect pri-miRNA and pre-miRNA processing and subsequent mature miRNA levels. For example, Wu et al. indicated that polymorphisms in the pri-let-7e result in decreased expression levels of mature miRNA [40]. Second, SNPs located within the seed site of miRNA can strengthen or decrease the interaction between the miRNA and target mRNA. For instance, Hu et al. reported that rs11614913 polymorphism in miR-196a2 could affect the miRNA-mRNA binding activity and be remarkably correlated with lung cancer survival [16]. Our bioinformatics findings indicated that rs2620381 polymorphism might affect miRNA function through both mechanisms. RNAfold web server output showed that the structural stability of miR-627 decreased in the presence of variant allele (ΔG increased from −56.40 kcal/mol to −51.66 kcal/mol). Moreover, miRmap web tool output indicated that the binding affinity of miR-627 to its mRNA targets reduced in the presence of variant allele.

Despite modest power, we have robustly supported the hypothesis that the A > C SNP in miR-627 rs2620381 is linked to gastric cancer, but cannot suggest this is causative. With a larger sample size, the borderline ($p = 0.06$) link between the AC genotype in the co-dominant model may become significant. We speculate this SNP predicts the risk of the development of this cancer, a finding that can only be determined with prospective studies. Nevertheless, this work represents an advance in biomedical science because it reports that the rs2620381 SNP in miR-627 is linked with gastric adenocarcinoma, and accordingly may be a risk factor.

Summary table

What is known about this topic:
- miR-627 plays significant roles in several biological pathways and diseases including cancer.
- miR-627 is significantly increased in gastric cancer.
- rs2620381 in miR-627 is not associated with the risk of lung cancer and esophageal squamous cell carcinoma.

What this paper adds:
- rs2620381 is significantly associated with gastric cancer
- There is no association between age, sex, tumour type, distant metastasis, and tumour stages and miR-627 polymorphism in gastric cancer patients.
- rs2620381 located in the seed site of miR-627 could affect the secondary structure of the miRNA and the miRNA-mRNA interaction

Acknowledgements

The study was conducted at the University of Guilan. The authors would like to thank all participants of the study.

Disclosure statement

No potential conflict of interest was reported by the authors.

ORCID

M Raad http://orcid.org/0000-0001-6530-3326
H Saeidi Saedi http://orcid.org/0000-0002-0910-7811

References

[1] Torre LA, Bray F, Siegel RL, et al. Global cancer statistics, 2012. CA Cancer J Clin. 2015;65:87–108. Available from: https://www.ncbi.nlm.nih.gov/pubmed/25651787www.gco.iarc.fr
[2] Wang F, Meng W, Wang B, et al. Helicobacter pylori-induced gastric inflammation and gastric cancer. Cancer Lett. 2014;345:196–202.
[3] Bouvard V, Baan R, Straif K, et al. A review of human carcinogens–part B: biological agents. Lancet Oncol. 2009;10:321.
[4] Xu Q, Liu J, Yuan Y. Comprehensive assessment of the association between miRNA polymorphisms and gastric cancer risk. Mutat Res. 2015;763:148–160.
[5] Berindan-Neagoe I, Monroig P Del C, Pasculli B, et al. MicroRNAome genome: a treasure for cancer diagnosis and therapy. CA Cancer J Clin. 2014;64:311–336.

[6] Di Leva G, Garofalo M, Croce CM. MicroRNAs in cancer. Annu Rev Pathol Mech Dis. 2014;9:287–314.
[7] Zhang B, Pan X, Cobb GP, et al. microRNAs as oncogenes and tumor suppressors. Dev Biol. 2007;302:1–12.
[8] Fabian MR, Sonenberg N. The mechanics of miRNA-mediated gene silencing: a look under the hood of miRISC. Nat Struct Mol Biol. 2012;19:586.
[9] Lewis BP, Burge CB, Bartel DP. Conserved seed pairing, often flanked by adenosines, indicates that thousands of human genes are microRNA targets. Cell. 2005;120:15–20.
[10] Waller P, Blann AD. Non-coding RNAs–A primer for the laboratory scientist. Br J Biomed Sci. 2019;74:157–165.
[11] Su Z-X, Zhao J, Rong Z-H, et al. Diagnostic and prognostic value of circulating miR-18a in the plasma of patients with gastric cancer. Tumor Biol. 2014;35:12119–12125.
[12] Shin VY, Ng EKO, Chan VW, et al. A three-miRNA signature as promising non-invasive diagnostic marker for gastric cancer. Mol Cancer. 2015;14:202.
[13] Chin LJ, Ratner E, Leng S, et al. A SNP in a let-7 microRNA complementary site in the KRAS 3′ untranslated region increases non–small cell lung cancer risk. Cancer Res. 2008;68:8535–8540.
[14] Kruglyak L, Nickerson DA. Variation is the spice of life. Nat Genet. 2001;27:234.
[15] Sun G, Yan J, Noltner K, et al. SNPs in human miRNA genes affect biogenesis and function. Rna. 2009;15:1640–1651.
[16] Hu Z, Chen J, Tian T, et al. Genetic variants of miRNA sequences and non–small cell lung cancer survival. J Clin Invest. 2008;118:2600–2608.
[17] Wei Y, Li L, Gao J. The association between two common polymorphisms (miR-146a rs2910164 and miR-196a2 rs11614913) and susceptibility to gastric cancer: A meta-analysis. Cancer Biomarkers. 2015;15:235–248.
[18] Xu L, Tang W. The associations of nucleotide polymorphisms in mir-196a2, mir-146a, mir-149 with lung cancer risk. Cancer Biomarkers. 2015;15:57–63.
[19] Kim WH, Min KT, Jeon YJ, et al. Association study of microRNA polymorphisms with hepatocellular carcinoma in Korean population. Gene. 2012;504:92–97.
[20] Schmit SL, Gollub J, Shapero MH, et al. MicroRNA polymorphisms and risk of colorectal cancer. Cancer Epidemiol Prev Biomarkers. 2014;24:65–72.
[21] Gao L-B, Bai P, Pan X-M, et al. The association between two polymorphisms in pre-miRNAs and breast cancer risk: a meta-analysis. Breast Cancer Res Treat. 2011;125:571–574.
[22] Padi SKR, Zhang Q, Rustum YM, et al. MicroRNA-627 mediates the epigenetic mechanisms of vitamin D to suppress proliferation of human colorectal cancer cells and growth of xenograft tumors in mice. Gastroenterology. 2013;145:437–446.
[23] Griffiths-Jones S, Grocock RJ, Van Dongen S, et al. miRBase: microRNA sequences, targets and gene nomenclature. Nucleic Acids Res. 2006;34:D140–D144.
[24] Sherry ST, Ward M-H, Kholodov M, et al. dbSNP: the NCBI database of genetic variation. Nucleic Acids Res. 2001;29:308–311.
[25] Gong J, Liu C, W L, et al. An update of miRNASNP database for better SNP selection by GWAS data, miRNA expression and online tools. Database. 2015;2015:bav029.
[26] Gruber AR, Lorenz R, Bernhart SH, et al. The vienna RNA websuite. Nucleic Acids Res. 2008;36:W70–4.
[27] Dweep H, Gretz N. miRWalk2. 0: a comprehensive atlas of microRNA-target interactions. Nat Methods. 2015;12:697.
[28] Chou C-H, Shrestha S, Yang C-D, et al. miRTarBase update 2018: a resource for experimentally validated microRNA-target interactions. Nucleic Acids Res. 2017;46:D296–302.
[29] Vejnar CE, Zdobnov EM. MiRmap: comprehensive prediction of microRNA target repression strength. Nucleic Acids Res. 2012;40:11673–11683.
[30] Lauren P. The two histological main types of gastric carcinoma: diffuse and so-called intestinal-type carcinoma: an attempt at a histo-clinical classification. Acta Pathol Microbiol Scand. 1965;64:31–49.
[31] Belgardt B-F, Ahmed K, Spranger M, et al. The microRNA-200 family regulates pancreatic beta cell survival in type 2 diabetes. Nat Med. 2015;21:619.
[32] Chan JA, Krichevsky AM, Kosik KS. MicroRNA-21 is an antiapoptotic factor in human glioblastoma cells. Cancer Res. 2005;65:6029–6033.
[33] Brennecke J, Hipfner DR, Stark A, et al. bantam encodes a developmentally regulated microRNA that controls cell proliferation and regulates the proapoptotic gene hid in Drosophila. Cell. 2003;113:25–36.
[34] Iliopoulos D, Jaeger SA, Hirsch HA, et al. STAT3 activation of miR-21 and miR-181b-1 via PTEN and CYLD are part of the epigenetic switch linking inflammation to cancer. Mol Cell. 2010;39:493–506.
[35] Li D, Zhu G, Di H, et al. Associations between genetic variants located in mature microRNAs and risk of lung cancer. Oncotarget. 2016;7:41715.
[36] Zhang P, Wang J, Lu T, et al. miR-449b rs10061133 and miR-4293 rs12220909 polymorphisms are associated with decreased esophageal squamous cell carcinoma in a Chinese population. Tumor Biol. 2015;36:8789–8795.
[37] Sun M, Zhang Q, Yang X, et al. Vitamin D enhances the efficacy of irinotecan through miR-627-mediated inhibition of intratumoral drug metabolism. Mol Cancer Ther. 2016;15:2086–2095.
[38] Chen F, Liu M, Yu Y, et al. LINC00958 regulated miR-627-5p/YBX2 axis to facilitate cell proliferation and migration in oral squamous cell carcinoma. Cancer Biol Ther. 2019;20:1270–1280.
[39] Cao J, Luo C, Yan R, et al. rs15869 at miRNA binding site in BRCA2 is associated with breast cancer susceptibility. Med Oncol. 2016;33:135.
[40] Wu M, Jolicoeur N, Li Z, et al. Genetic variations of microRNAs in human cancer and their effects on the expression of miRNAs. Carcinogenesis. 2008;29:1710–1716.