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Original Article

Expression of miR-199a in gastric cancer tissues and serum and its clinical significance

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Abstract: Objective: To evaluate the diagnostic values of miR-199a in gastric cancer tissues and serum by analyzing its expressions in gastric cancer tissues and serum. Methods: A total of 42 patients with gastric cancer were collected as the experimental group, and 60 healthy individuals of similar age as the control group. The specimens were newly collected from the patients after radical gastrectomy. The cancer tissues and distal tissues (over 5 cm from the cancer tissues) of patients were harvested 30 minutes postoperatively for fast freezing, which confirmed gastric cancer, and the tissues which had been cut were placed in liquid nitrogen until use. The peripheral blood (5 ml) was collected from patients at 1 week pre- and post-operatively, and peripheral blood of the same volume was also collected from healthy individuals (control group). The expressions of miR-126 and miR-199a in tissues and serum were detected by real-time quantitative PCR. A total of 5 ml of peripheral blood was collected from the patients approximately at 1 week pre- and post-operatively for detection of standardized cancer carcinoembryonic antigen (CEA), with the reference value at 0 to 10 ng/L. Results: The results of RT-PCR showed that the relative expression level of miR-199a in cancer tissues (12.04 ± 2.27) was higher than that in the distant cancer tissues (control group, 2.12 ± 1.11), indicating that there was significant difference between the two groups ($t = 3.908$, $P<0.05$); the expression of miR-199a in cancer tissues was associated with the clinical stage of the tumor, depth of invasion, and lymph node metastasis. The relative expression level of miR-199a in serum (30.04 ± 5.04) before operation was higher than that (14.03 ± 6.02) after operation ($t = 6.778$, $P<0.05$); the relative expression in serum before operation was higher than that in healthy controls (5.81 ± 4.67, $t = 7.684$, $P<0.05$), indicating significant difference between the two groups. The expression of miR-199a in serum was associated with the clinical stage of tumors, depth of invasion, and lymph node metastasis. With regard to the diagnosis of gastric cancer, the AUC of the diagnostic efficiency ROC curve of miR-199a in gastric cancer tissues and serum was 0.862 and 0.918, and the sensitivity and the specificity were 92.9% and 71.4%, respectively. The AUC of the diagnostic efficiency ROC curve of CEA was 0.537. Pearson correlation analysis showed that the correlation coefficient r of the expression of miR-199a in cancer tissues and preoperative serum was 0.65 ($P<0.01$), suggestive of their positive correlation. Conclusions: The expressions of miR-199a in gastric cancer tissues and serum are up-regulated, and associated with the development and progression of gastric cancer. The expression of serum miR-199a can serve as a marker to evaluate the severity of gastric cancer. The expression of miR-199a is superior to CEA in terms of diagnostic values for gastric cancer. There is a positive correlation in miR-199a expression between tissues and serum, and miR-199a is expected to be a valuable biomarker for the diagnosis of early gastric cancer and monitoring of gastric cancer recurrence.

Keywords: miR-199a, cancer tissue, serum, expression, real-time quantitative PCR, diagnosis, tumor markers

Introduction

Gastric cancer is one of the common malignancies with the incidence and mortality ranking top three malignancies, and China has high incidence of gastric cancer throughout the world. Gastric cancer has occult early symptoms, with the vast majority of newly diagnosed patients already at the middle or late stage of gastric cancer, infiltration of cancer tissues into the muscle layer or even the serosa, and a five-year survival of less than 5% [1]. A large number of patients succumb to postoperative recurrence and metastasis. At present, the diagnosis of gastric cancer is mainly gastroscopic biopsy followed by pathological examinations.

As an invasive examination, gastroscopy cannot serve as a routine means for screening and its prevalence is very low. The diagnosis of early gastric cancer in China is less than 10% [2]. Carcinoembryonic antigen (CEA) is a broad-spectrum tumor marker with poor sensitivity and specificity for the diagnosis of gastric cancer [3]. Therefore, it is of great significance to seek a noninvasive early biological marker for the diagnosis of gastric cancer. The MicroRNA (mi RNA) is a type of endogenous single-stranded noncoding small RNA with regulatory function about 22 nt in length in eukaryotic cells. By binding to 3'-untranslated Regions of the target mRNA, it inhibits the post-transcriptional translation process of the target mRNA. It is involved in the whole process, including development, progression, invasion, metastasis, proliferation and apoptosis, whereby it serves as an oncogene or tumor suppressor gene. MiR-199a is differentially expressed in tumors, with its expression in the gastric cancer tissues and serum not very clear. Mi RNA is relatively stable in the body fluid [4], and is free from repeated freezing and thawing, and changes in PH values, etc. Herein, we detected the expressions of miR-199a in gastric carcinoma and serum by real-time quantitative PCR, analyzed the relationship between miR-199a expressions and clinicopathological parameters, and compared the difference between miR-199a as a marker of gastric cancer and CEA. Furthermore, we analyzed the correlation between miR-199a expressions in tissues and serum, and explored the diagnostic values of miR-199a in tissues and serum as a tumor marker of gastric cancer.

Materials and methods

Materials

Source of patients and collection of clinical data: A total of 42 gastric cancer patients with complete data who had been hospitalized in the gastrointestinal surgery department of No. 2 Hospital Affiliated to Nantong University from June 2006 to December 2016 were enrolled, including 27 male patients aged 47 to 78 years and 15 females aged 33 years to 83 years. Patients were enrolled if they met the following criteria: 1. They had undergone preoperative gastroscopy, whereby they had pathologically confirmed gastric cancer; 2. All patients were newly diagnosed patients, and had no tumors of important organs, such as the liver, the kidney and the digestive system; 3. They had not undergone preoperative radiotherapy, chemotherapy or other related treatment; 4. Routine postoperative pathological tissue sections for the patients confirmed gastric cancer once again. Gastric cancer was staged in accordance with the TNM staging standards developed by the Union for International Cancer Control (UICC) in 2016, based on which there were 30 patients with stage I and II gastric cancer, 12 with stage III and IV gastric cancer, 15 with T1 gastric cancer, 22 with T2 gastric cancer, 7 with T3 gastric cancer, 8 with T4 gastric cancer, 15 with lymph node metastasis, and 27 with no lymph node metastasis. The number of patients with high plus medium and low differentiation was 9 and 33, respectively, and the number of those with a tumor diameter ≥ and <5 cm was 14 and 28, respectively. During the same period, a total of 60 healthy subjects of similar age and gender who had undergone physical examinations in the physical examination center of Nantong University Hospital were also enrolled, including 37 males and 23 females.

Collection of experimental samples: Two cancer tissues and two corresponding distal cancer tissues (control group, >5 cm from the cancer tissues), 1 × 1.5 cm^2 in size and of uniform thickness (2~3 mm), were divided from fresh tissue specimens of gastric cancer postoperatively 30 minutes after they had been taken out of the patients. The tissues were marked, and placed in RNase-free centrifuge tubes. A proportion of cancer tissues and distal cancer tissues (control group) that had been cut were made into fast frozen pathology sections and conventional paraffin pathology sections before HE staining, and observed under a light microscope, which confirmed that cancer tissues accounted for above 60% of all tissues in the field in the gastric cancer group, and that no tumor tissues were found in the distal cancer group (control group). Another proportion of the tissues that had been cut were frozen quickly, and then stored in a refrigerator at -70°C until use. A total of 5 ml of peripheral blood was collected from the patients approximately 1 week pre- and postoperatively before being sent to the clinical laboratory department for detection of standardized CEA. Peripheral blood (5 ml) was collected from patients about 1 week pre- and postoperatively, and peripheral blood of the same volume was also collected from

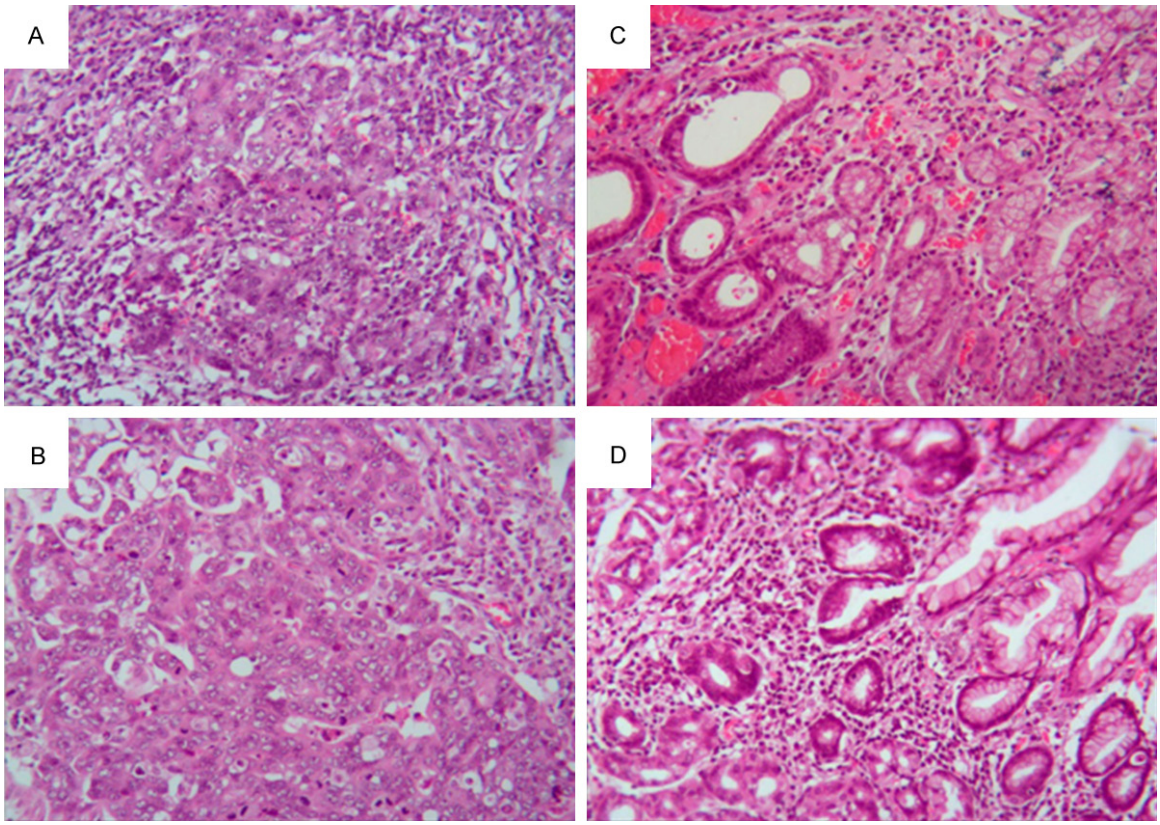

Figure 1. Pathology and histology of cancer tissues and distal cancer tissues. (A and B) represent cancer tissues, (C and D) indicate distant cancer tissues (control group, H-E × 200).

healthy individuals (control group). Subsequently, the blood was placed in the pro-coagulation tube, and centrifuged, within 30 minutes, at 3500 rpm for 10 min. The serum was placed in a 2.0 ml RNase-free centrifuge tube, and then placed in a -70°C refrigerator until use.

Reagents and instruments: The MiRcute miRNA extraction kit (centrifugal plate type), the miRcute mi RNA extraction and isolation kit (centrifugal column type), the miRcute enhanced miRNA cDNA first-chain synthesis kit and the miRcute enhanced miRNA fluorescence quantitative detection kit were all purchased from Beijing Tiangen biochemical company. The ultramicro total spectrophotometer OneDrop1000 and the real-time PCR instrument were purchased from Shanghai Noruo Jing Biotechnology Co., Ltd. and StepOne Plus, respectively.

Detection of expressions of miR-199a using qRT-PCR

Extraction of total RNA and reverse transcription: The total RNA was extracted from the cancer tissues and the distal cancer tissues (control group) by the miRcute mi RNA extraction kit, and the serum total RNA was extracted using the miRcute serum/plasma miRNA extraction kit. The concentration and purity of RNA were detected using the ultra-micro spectrophotometer, with the value of the relatively pure RNA A260/A280 of 1.8 to 2.0. MiRNA reverse transcription was performed in accordance with the instructions of the miRNA reverse transcription kit by Genomics Biochemicals. The total reaction system was 20 μl, including the total RNA (5 μl of the tissue extract or 8 μl of the serum extract), 10 μl of the 2 × mi RNA RT Reaction Buffer, 2 μl of mi RNA RT Enzyme Mix, and supplementation of RNase-Free H_2O until 20 μl. Reverse transcription was performed at 42°C for 60 min, with miRNA in the A tail reaction and the reverse transcription reaction. The enzyme inactivation reaction was conducted at 95°C for 3min. The synthesized cDNA reaction solution was stored at -20°C.

qRT-PCR: The synthesized cDNA reaction solution was diluted at the ratio of 50:1, and the

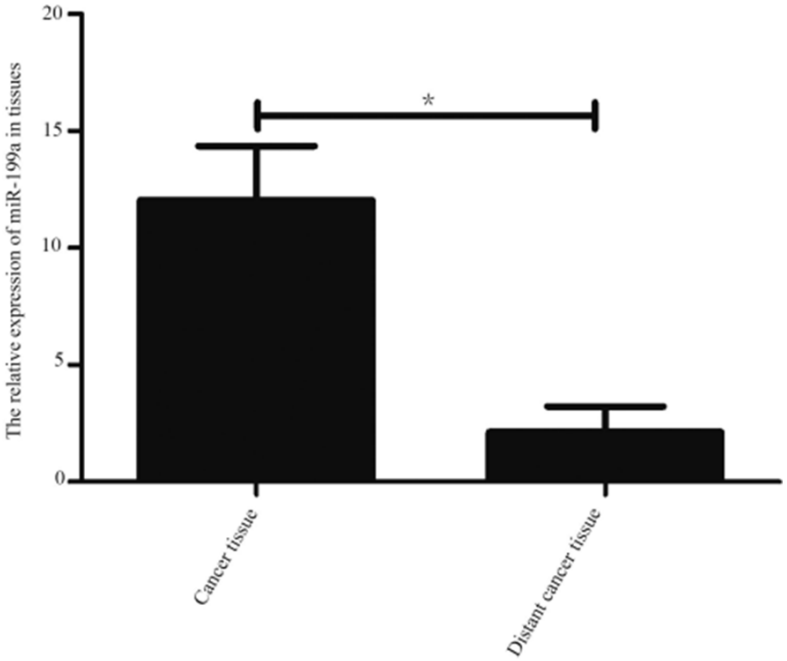

Figure 2. Expressions of miR-199a in cancer tissues and distal cancer tissues in 42 patients with gastric cancer.

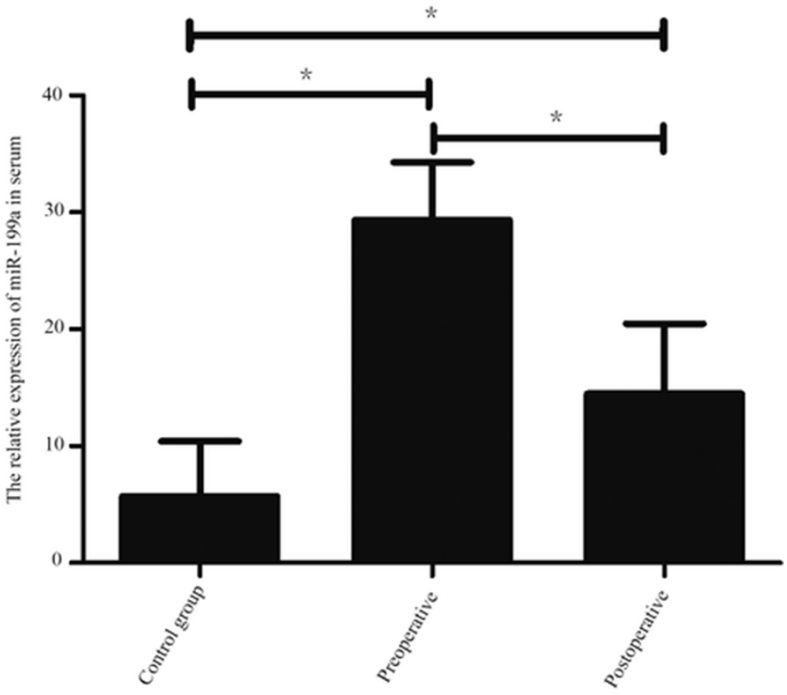

Figure 3. The expression levels of serum miR-199a of 42 patients with gastric cancer pre- and postoperatively and the serum miR-199a of 42 healthy controls.

miR-199a was detected in accordance with the instructions of the miRcute enhanced miRNA quantitative assay kit. The PCR system was 20 μl, including 2 × miRcute Plus mi RNA Premix (10 μl), Forward Primer (0.4 μl), Reverse Primer (0.4 μl), first strand cDNA of mi RNA (2 μl), 50 × ROX Reference Dye (2 μl), and supplementation of RNase-Free H_2O until 20 μl. The tissue reaction procedures were as follows: 95°C, 15 min for one cycle; 94°C, 20 sec; 60°C, 34 sec for 45 cycles. The serum reaction procedures were as follows: 95°C, 15 min for one cycle; 94°C, 20 sec; 65°C, 30 sec; 72°C, 34 sec for 5 cycles; 94°C, 20 sec; 60°C, 34 sec for 45 cycles. Experiments for all the samples were repeated in triplicate to obtain the Ct values, and the relative expressions of miRNA199a in cancer tissues, distal cancer tissues (control group) and serum were calculated using the formula $2^{-\Delta\Delta Ct}$.

Statistical treatment

The Graphpad Prism 5.0 software was adopted to process the charts, and all data were statistically analyzed using the SPSS Statistics 19.0 statistical software. The values between the two groups and multiple groups were compared using *t*-test or one-way analysis of variance. The measurement data were expressed as mean ± standard deviation (SD), and the count data analysis was performed using the chi-square test. The diagnostic values of miR-199a for the detection of gastric cancer were represented by ROC curve, and Pearson correlation analysis was used to analyze the correlation between groups. $P<0.05$ suggested that the difference was statistically significant.

Table 1. The expression of miR-199a in tissues with different groups of gastric cancer

Clinicopathological characteristics	N	Relative expression levels ($2^{-\Delta\Delta Ct}$)	t/F	p
Sex			0.948	0.443
Male	27	8.05 ± 8.07		
Female	15	19.692 ± 29.47		
Age			0.532	0.614
≥60	31	10.012 ± 1.26		
<60	11	11.27 ± 2.63		
Tumor location			0.625	0.541
Gastric antrum and lesser curvature	32	11.54 ± 2.70		
Pylorus and greater curvature	5	12.30 ± 2.20		
Cardia and fundus	5	12.306 ± 2.20		
Tumor size			1.387	0.718
≥5 cm	14	17.73 ± 2.01		
<5 cm	28	9.44 ± 1.84		
Clinical stages			4.834	0.004
Stages I and II	30	10.6 ± 3.25		
Stages III and IV	12	16.47 ± 1.36		
Depth of invasion			5.841	0.003
T1	5	15.02 ± 1.79		
T2	22	15.29 ± 1.84		
T3	7	16.30 ± 219		
T4	8	19.55 ± 3.36		
Degree of differentiation			2.918	0.077
Middle and high differentiation	9	5.28 ± 4.29		
Low differentiation	33	14.01 ± 2.24		
Lymphatic metastasis			7.729	0.000
Yes	15	13.72 ± 3.20		
No	27	6.99 ± 2.69		

Results

Pathological analysis of gastric cancer tissues and distal cancer tissues

The specimens of gastric cancer tissues and distal cancer tissues were made into frozen sections and paraffin-embedded sections before HE staining. It was confirmed under the light microscopy that the cancer tissues accounted for over 60% of the total tissues in the field in the gastric cancer group, and that there were no cancer tissues in the distal cancer group (control group). See **Figure 1**.

Expressions of miR-199a in gastric cancer tissues and serum

The expression level of miR-199a was (12.04 ± 2.27) in the tissue of gastric cancer patients, which was much higher than that in the distant cancer tissues (control group, 2.12 ± 1.11, t = 3.908, P<0.05), and the difference was statistically significant. See **Figure 2**.

The expression level of miR-199a in the serum of gastric cancer patients (30.04 ± 5.04) was significantly higher than that after operation (14.03 ± 6.02, t = 6.778, P<0.05), and the serum miR-199a expression level after operation was higher than that in the healthy control group (5.81 ± 4.67, t = 7.684, P<0.05). The difference was statistically significant between the two groups. See **Figure 3**.

Relationship between miR-199a expressions and clinicopathological characteristics

The expressions of miR-199a in gastric cancer patients in different clinicopathological groups are shown in **Table 1**. The expression of miR-199a was associated with the clinical stage, depth of invasion and lymphatic metastasis of tumors, and the expression of miR-199a -199a was increased as the increased depth of invasion and progression of stages. The difference between the groups was statistically significant, and P<0.01. However, there was no significant difference between the expressions and patients' age, sex, tumor location, tumor size and differentiation degree, with P> 0.05.

As shown in **Table 2**, the expression of miR-199a in the serum of patients with gastric cancer was associated with the clinical stage and depth of invasion of the cancer. The expression of serum miR-199a was increased with the rise in the depth of invasion and progression of stages, and the difference was statistically significant (P<0.01). However, there was no significant difference between the expression and patients' age, sex, tumor location, tumor size,

Table 2. The expression of miR-199a in preoperative serum among different groups of gastric cancer

Clinicopathological characteristics	N	Relative expression levels ($2^{-\Delta\Delta Ct}$)	t/F	p
Sex			0.862	0.224
Male	27	32.00 ± 4.98		
Female	15	28.12 ± 6.12		
Age			0.977	0.123
≥60	31	29.82 ± 5.39		
<60	11	27.64 ± 7.23		
Tumor location			0.211	0.811
Gastric antrum and lesser curvature	32	32.55 ± 4.12		
Pylorus and greater curvature	5	30.20 ± 3.12		
Cardia and fundus	5	30.86 ± 4.98		
Tumor size			0.939	0.153
≥5 cm	14	32.66 ± 5.2		
<5 cm	28	29.61 ± 6.21		
Clinical stages			4.482	0.004
Stages I and II	30	19.95 ± 5.76		
Stages III and IV	12	30.20 ± 7.84		
Depth of invasion			4.123	0.004
T1	5	29.66 ± 4.45		
T2	22	31.65 ± 5.12		
T3	7	32.01 ± 2.83		
T4	8	34.12 ± 4.82		
Degree of differentiation			1.522	0.056
Middle and high differentiation	9	35.67 ± 4.5		
Low differentiation	33	29.25 ± 2.94		
Lymphatic metastasis			1.317	0.062
Yes	15	25.52 ± 7.94		
No	27	34.02 ± 5.76		

differentiation degree and lymphatic metastasis, with P>0.05.

Diagnostic efficiency of miR-199a and CEA in tissues and serum of gastric cancer patients

The diagnostic values of miR-199a and CEA in tissues and serum were expressed as the ROC curve and AUC. See **Figures 4**-**6**.

Analysis of the correlation between miRNA199a expression in gastric cancer tissues and serum

Pearson correlation analysis was adopted, indicating that the correlation coefficient r for the relative expression level of miR-199a in cancer tissues and serum in gastric cancer patients was 0.65, indicating that they were positively correlated with each other (P<0.01). See **Figure 7**.

Discussion

Studies on miR-199a, two precursors of which are derived from chromosome 1 and 19, have gradually attracted eyes. It degrades into miR-199a-3p, miR-199a-5p and miR-199a-3p in the presence of Dicer enzyme, and the miR-199a-3p is located within the intron 16 of the DNM2 gene at p13.2 of chromosome 19. Part of miR-199a is down-regulated as a tumor suppressor gene and partly up-regulated as oncogene, and abnormal expressions of miR-199a are closely associated with the development and progression of tumors [5]. Circulatory miRNAs include plasma miRNAs and serum miRNAs. Studies have shown that the contents of miRNAs are roughly equal in plasma and serum [6, 7], and that circulatory miRNAs are mainly released by damaged, inflammatory or necrotic cells [8]. They bind with high-density lipoprotein, RNA binding protein (AGO2) and nucleolus phosphate protein 1 to form a complex and are released after activation [9]; they are activated and secreted by microvesicles (MV), exosomes and apoptotic bodies [10]. Easy collection of clinical blood samples, basically noninvasive operations, convenient detection and good acceptance by patients make miRNA a possible molecular marker of tumors theoretically and clinically, while studies on miRNA as a tumor marker remain to be further carried out. The expressions of miRNA are also detected in gastric cancer tissues, serum and gastric juice, while the expressions of miR-199a in cancer tissues are rarely reported.

Diagnostic values of serum miR-199a in patients with gastric cancer

MiR-199a is differentially expressed in various tumors, which is lowly expressed in hepatocel-

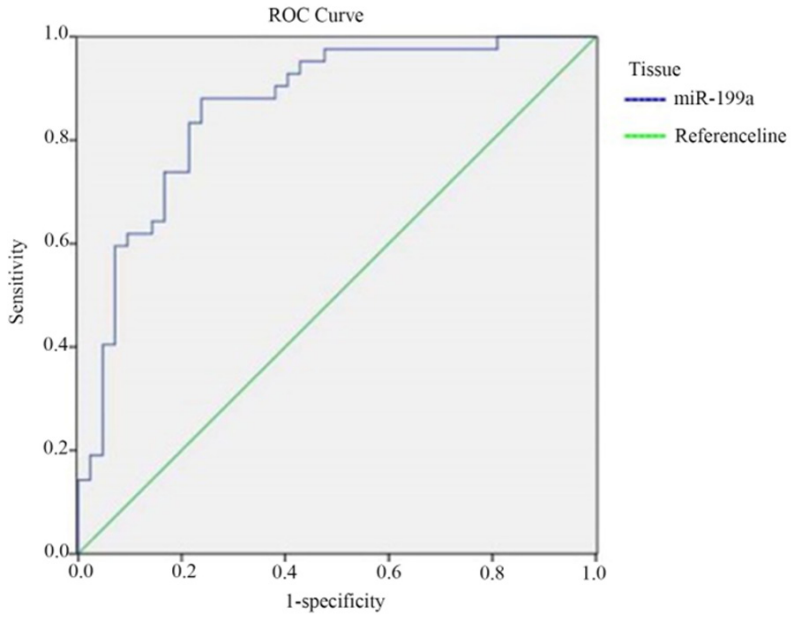

Figure 4. Diagnostic efficiency of miR-199a in gastric cancer tissues. AUC = 0.862, (95% CI: 0.782~0.942). When the cutoff value was at 1.3, the sensitivity and specificity were 88.1% and 76.2%, respectively.

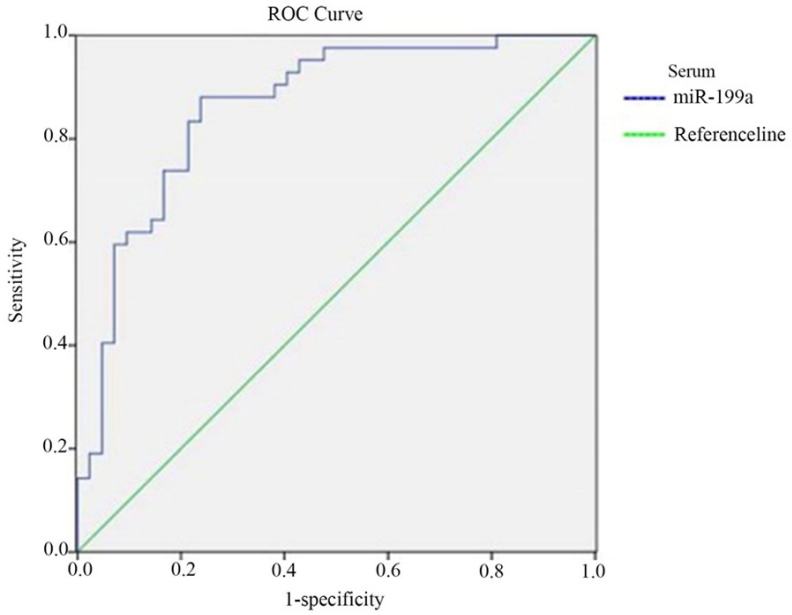

Figure 5. Diagnostic efficiency of preoperative serum miR-199a in patients with gastric cancer. AUC = 0.918, (95% CI: 0.863~0.974). When the cutoff value was at 7.18, the sensitivity and specificity were 92.9% and 71.4%, respectively.

lular carcinoma and colorectal cancer while highly expressed in esophageal cancer, cholangiocarcinoma and pancreatic cancer [11-13]. Brenner *et al.*, however, found that miR-199a was highly expressed in gastric cancer [14]. This study showed that the relative expression level of miR-199a in 42 patients with gastric cancer was 12.04 ± 2.27, much higher than that in the distant cancer (control group, 2.12 ± 1.11), consistent with the relevant reports. The preoperative relative expression level of serum miR-199a (30.04 ± 5.04) was significantly higher than that after operation (14.03 ± 6.02) and higher than that in the healthy control group (5.81 ± 4.67). The relative expression level of serum miR-199a after operation was higher than that in healthy control group, suggesting that serum miR-199a might also represent a new noninvasive means for the detection of gastric cancer. Studies showed that target genes of miR-199a included AXL, AKT, ZHX1, c-Met, PAK4 and HIF-1α, etc. In liver cancer, miR-199a acted on target HIF-1α directly to inhibit its expression, thereby inhibiting tumor proliferation [15]. In gastric cancer, miR-199a-3p promotes the apoptosis of gastric cancer cells by inhibiting ZHX1 [16]. In order to better evaluate the clinical diagnostic values of miRNA-126 and miR-199a, the Receiver Operator Characteristic Curve (ROC curve) was drawn in this study based on the detection results in gastric cancer tissues and serum, with the value of AUC between 0.5 and 1. The value closer to 1 indicated better diagnostic effects, and the value between 0.5 to 0.7, 0.7 to 0.9 and above 0.9 denoted lower accuracy, certain accuracy and high accuracy. The maximum cutoff point for the Youden index is the optimal critical value. The results showed that when the miR-199a was diagnosed in gas-

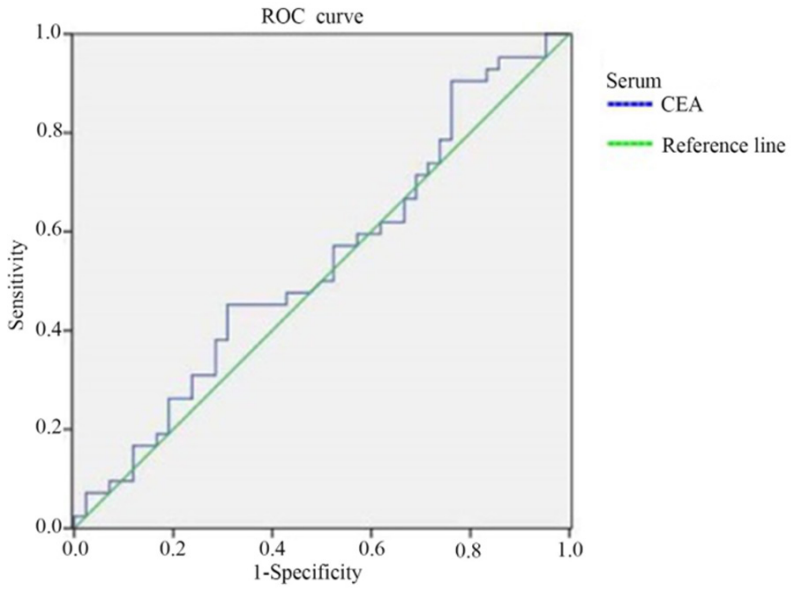

Figure 6. Diagnostic efficiency of serum CEA in patients with gastric cancer. AUC = 0.537.

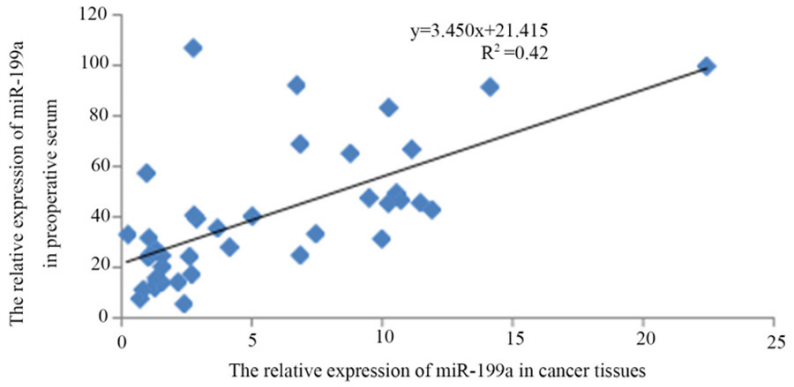

Figure 7. Correlation between miR-199a expression in tissues and serum.

tric cancer in the 42 fresh gastric specimen tissues, AUC was 0.862 in the tissues, and the sensitivity and specificity were 88.1% and 76.2%, respectively, when the threshold was set at 1.30. The serum AUC was 0.918, and the sensitivity and the specificity were 92.9% and 71.4%, respectively, when the critical value was set at 7.18. CEA, the most common tumor marker whose gene is located at chromosome 19, is a type of acidic glycoprotein. It is widely found in digestive system tumors and lung cancer with the sensitivity to malignant digestive system tumors of 50 to 70% [17], and it is of great significance for the detection of gastric cancer, the judgment of curative effects and prognosis evaluation [18]. However, its specificity is poor and vulnerable to smoking, infection and other influences. The normal reference value of CEA in this study was 0 to 10 ng/L, and AUC was only 0.537 in the diagnosis of gastric cancer. The AUC of miR-199a was about 0.9 in tissues and serum during the diagnosis of gastric cancer with over 70% high sensitivity and specificity, demonstrating significant diagnostic values. The diagnostic value of miR-199a was superior to that of CEA. MiR-199a met the characteristics of tumor markers, such as high sensitivity and specificity, Moreover, the expression levels are associated with tumor sizes and clinical stages. Therefore, serum miR-199a can be used as a noninvasive method for the diagnosis of gastric cancer and monitoring of gastric cancer recurrence.

The expression of serum miR-199a in patients with gastric cancer and its values in disease evaluation and prognosis

Currently, studies have shown that there is a certain correlation between the expression level of miR-199a in tumor tissues and serum and the clinicopathological parameters. Yin *et al.* [19] found that miR-199a had certain diagnostic value in the prognosis of patients with hepatocellular carcinoma, and that the expression level of miR-199a in plasma of patients with hepatocellular carcinoma was positively correlated with their survival after operation. In this study, real-time quantitative PCR was employed to detect the expression levels of miR-199a in gastric cancer tissues, distal cancer tissues and serum of patients before and after surgery, and the relationship between the expressions of miR-199a and clinicopathological parameters, including patients' age, sex, tumor location, tumor size, depth of invasion, staging,

degree of differentiation and metastasis was analyzed. The results showed that the miR-199a expression level was not significantly associated with patients' age, gender, and tumor location. The relative expression level of miR-199a in serum of patients with gastric cancer before operation was related to the clinical stage and depth of tumor invasion. The relative expression levels of miR-199a in different stages were as follows: stages I and II, 19.95 ± 5.76; stages III and IV, 30.20 ± 7.84. Besides, the relative expression levels of miR-199a in different depth of infiltration were as follows: T1, 29.66 ± 4.45; T2, 31.65 ± 5.12; T3, 32.01 ± 2.83; and T4, 34.12 ± 4.82. The differences between the two groups were statistically significant (P<0.01). Furthermore, the relative expression level of miR-199a in serum was increased with the progression of stages and the depth of invasion, consistent with the relative expression level of miR-199a in the tissue. The relative expression level of miR-199a in tissues was also correlated with lymphatic metastasis, with the relative expressions of miR-199a in lymphatic metastasis group and the non-lymphatic metastasis group being 13.72 ± 3.20 and 6.99 ± 2.69, respectively, which might be related to patients' prognosis. In this study, serum of patients was collected at one week postoperatively as controls, and the serum can be collected again one month or later after operation to dynamically observe the changes in the expression levels in the serum, thereby inferring the recurrence of gastric cancer patients or the time window of metastasis. Therefore, miR-199a may play an important role in the development, progression and prognosis of gastric cancer, and serum miR-199a can be used as a biomarker to evaluate the severity and prognosis of patients with gastric cancer.

Correlation between the expressions of miR-199a in gastric cancer tissues and serum

Weber *et al.* [20] demonstrated that miRNAs could be detected within body fluids, such as blood, cerebrospinal fluid, sputum, pleural effusion and alveolar lavage fluid, as well as in benign gastric diseases such as gastritis [21]. Its good stability was determined by its special presence within different body fluids. miRNAs within tissues and cells enter the humoral circulation by active secretion, lysis and release of circulatory cells and apoptotic necrotic cells, while miRNAs in tumor cells are transported into circulation via tumor-associated bodies [22]. The results of this experiment showed that the correlation efficient of the relative expressions level of preoperative miR-199a in cancer tissues and serum among gastric cancer patients was r = 0.65 (P<0.01), indicating that both were positively correlated with each other. Therefore, we suspected that miRNAs in serum were caused by the release of gastric cancer cells into the blood. In addition, the trend of changes in serum miR-199a expressions was consistent with that in the corresponding cancer tissues, showing good prospects for the early diagnosis of gastric cancer and evaluation of the disease. However, due to the limited number of fresh specimens and the low and moderate correlation between the expressions of miR-199a in tissues and serum, further studies regarding the correlation with a larger sample size remain to be carried out. The relationship with survival during the next 3 to 5 years remains to be further investigated.

To sum up, we quantitatively analyzed the relationship between the expressions of miR-199a in gastric cancer tissues and serum and the clinicopathological features, which was conducive to monitoring of the disease and evaluation of its prognosis. Besides, the diagnostic values of both as biomarkers of gastric cancer were analyzed and the correlation between the expressions of miR-199a in gastric cancer tissues and serum was discussed. These jobs are significant for exploration of the mechanisms underlying the development and progression of gastric cancer.

Disclosure of conflict of interest

None.

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