SPANDIDOS PUBLICATIONS

ONCOLOGY LETTERS 20: 2456-2464, 2020

Diagnostic value and prognostic significance of LI-cadherin and miR-378e in colorectal cancer

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Received December 14, 2018; Accepted December 3, 2019

DOI: 10.3892/ol.2020.11755

Abstract. Expression levels of LI-cadherin and miR-378e in the serum of patients with colorectal cancer, and the diagnostic value and prognostic significance in colorectal cancer were investigated. A total of 110 patients who were diagnosed with colorectal cancer in Weihai Central Hospital, from January 2012 to November 2014, were selected and enrolled in the experimental group, and 90 healthy subjects who underwent physical examination were enrolled in the control group. The expression level of miR-378e in serum was detected by reverse transcription-quantitative PCR and the expression of LI-cadherin in serum was detected by ELISA. ROC curves of LI-cadherin and miR-378e were drawn and the sensitivity and specificity of the diagnosis were estimated. The association of the expression levels of LI-cadherin and miR-378e with the survival of the patients was analyzed. LI-cadherin and miR-378e expression levels were significantly higher in the control group than those in the experimental group (P<0.001). LI-cadherin was significantly associated with the pathogenic site, the lymphatic metastasis, depth of infiltration, degree of differentiation and clinical stage (P<0.05). The sensitivity and specificity of the LI-cadherin combined with miR-378e detection were respectively 86 and 94%; the sensitivity of miR-378e detection was the highest, as well as the specificity of the combined detection. At the end of the follow-up period, the survival rates of the patients in the LI-cadherin high-expression group and miR-378e high-expression group were significantly higher than those in the low-expression groups (P<0.05). There was a significant positive correlation between the LI-cadherin and miR-378e expression levels in both the experimental and control group (r=0.5845 and 0.6356, respectively; P<0.05). In conclusion, LI-cadherin and miR-378e are expressed at low levels in colorectal cancer, suggesting that they have a good diagnostic value for colorectal cancer and can be used as biomarkers for colorectal cancer prognosis.

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Key words: colorectal cancer, LI-cadherin, miR-378e, diagnostic value, prognosis

Introduction

Colorectal cancer is the most common gastrointestinal cancer worldwide. The morbidity of colorectal cancer has increased over the past 20 years in most countries (1). The aging of population and the high-fat and low-fiber diet are the main reasons of colorectal cancer. The initial onset of colorectal cancer is insidious; in most cases colorectal cancer has no clinical symptoms and there are different degrees of delayed diagnosis (2). At present, surgery, chemotherapy and radiotherapy are the main treatments for colorectal cancer (3,4). Although the treatment methods are constantly improving, recurrence and metastasis still occur after the treatment of colorectal cancer by these methods. Thus, colorectal cancer still poses a threat to human health (5). Serum tumor markers, such as carcinoembryonic antigen and carbohydrate antigen 199, have greatly improved the diagnostic levels; however, they are only used in postoperative monitoring and they are not suitable for the early detection of colorectal cancer (6-8). Therefore, finding new tumor markers is important to improve the diagnosis and prognosis in colorectal cancer.

LI-cadherin was initially discovered in the liver and intestinal tract of rats (9). As a non-classical cadherin, LI-cadherin has its own unique structure. Unlike the traditional cadherins, which have five E-cadherin iterons, LI-cadherin has seven independent domains outside the cells and there are 20 amino acid residues in cytoplasm, indicating that there is less homology between LI-cadherin and other cadherins (10). The expression of LI-cadherin is closely associated with tumors related to the digestive system, and LI-cadherin expression is often associated with tumor prognosis (11,12). At present, there is a number of studies on the LI-cadherin expression in gastric, esophageal and pancreatic cancers (13). In recent years, LI-cadherin has been a hotspot in research; however, there are few studies on LI-cadherin in colorectal cancer.

MicroRNA (miRNA) is an endogenous, 20-23 nucleotide-long, non-coding, single-stranded RNA (14), which plays an important role in regulation after genes are transcribed, and plays a key role in the development of the organisms, the differentiation of cells, the cell signaling, the regulation of the expression of genes, and the occurrence and development of

malignant tumors (15). miRNAs are abnormally expressed in various malignant tumors. For example, the expression of miR-155 is upregulated in breast cancer (16), the expression of miR-221 is upregulated in pancreatic cancer (17), and the expression of miR-885-5p is downregulated in hepatoma (18). miRNA is considered to be involved in the occurrence and development of tumors, and thus, may be regarded as an oncogene or a tumor suppressor gene. Lu *et al* (19) have reported that the tissue source and differentiation status of various tumors can be more accurately reflected by the expression profile of miRNA. Therefore, the expression profile of miRNA can be used for the classification of various poorly differentiated tumors, indicating that miRNA is very important in the diagnosis of tumors and the prognosis of cancer. A study on miR-378e is rare in China and globally. A relevant study has shown that the expression of miR-378e is downregulated in colorectal cancer tissue, suggesting that miR-378e may be considered as a tumor suppressor gene and could be used as a potential molecular marker for colorectal cancer (20). However, currently there is no other study to verify this and the specific effect still needs to be further investigated.

The aim of the present study was to investigate the expression levels of LI-cadherin and miR-378e in the serum of patients with colorectal cancer, and to analyze the diagnostic value and prognostic significance of LI-cadherin and miR-378e in colorectal cancer.

Subjects and methods

General data. A total of 110 patients who were diagnosed with colorectal cancer in Weihai Central Hospital (Weihai, China), from January 2012 to November 2014, were selected and enrolled in the experimental group. There were 66 males and 44 females included in the experimental group, 62.13±9.21 years of age. In addition, there were 69 cases in stages A and B, and 41 cases in stages C and D.; and there were 45 cases with lymphatic metastasis, and 65 cases without lymphatic metastasis. At the same time, 90 healthy subjects who underwent physical examination were selected and enrolled in the control group. There were 52 males and 38 females included in the control group, 61.89±9.28 years of age.

Inclusion criteria: Patients who had complete clinicopathological data; patients who had not received neoadjuvant chemotherapy, radiotherapy or endocrinotherapy; patients who had completed some examinations within 2 weeks before the operation, including hepatorenal function, tumor markers, blood routine and urine routine examinations.

Exclusion criteria: Patients with other malignant tumors; patients with severe congenital heart disease; patients with severe organ lesion or severe organ disease; patients with autoimmune system disease; women in gestation or lactation period; patients who did not cooperate with the examinations; patients whose family refused to sign the informed consent form.

The study was approved by the Ethics Committee of the Weihai Central Hospital. All participants were informed in detail on the experiments of this research and had complete clinical data. Signed written informed consents were obtained from the participants of this study and/or their guardians.

Collection of serum. After the patients were diagnosed with colorectal cancer, 2 ml of peripheral blood were collected after having fasted in the morning. Blood samples were added into anticoagulation tubes for further analysis. In the control group, 2 ml of fasting venous blood were collected in the morning. After the blood was coagulated for 60 min (between 20 and 25˚C), centrifugation was carried out for 10 min at 4˚C at a speed of 1,006.2 x g. Next, the supernatant was collected and placed at -80˚C for further analysis. Repeated freezing and thawing were avoided as much as possible.

Experimental reagents and instruments. TRIzol® kit (Thermo Fisher Scientific, Inc.); DNase I (Shanghai Shenggong Biology Engineering Technology Service, Ltd.); cDNA reverse transcription kit (Takara Bio, Inc.); ultraviolet spectrophotometer (Shanghai Mepuda Instrument Co., Ltd.); SYBR Premix Ex Taq™ kit (Takara Bio, Inc.); quantitative PCR detector (ABI 7300; Shanghai Zhiyan Scientific Instrument Co., Ltd.); LI-cadherin enzyme-linked immunosorbent assay (ELISA) test kit (R&D Systems China Co., Ltd.); BS-1101 enzyme microplate reader (Beijing Linmao Technology Co., Ltd.).

Detection of miR-378e expression by reverse transcription-quantitative PCR. Total RNA in serum was extracted using TRIzol® reagent, according to the manufacturer's protocol. DNase I (RNase-free) was used to digest the template RNA in order to eliminate the contamination of genome DNA. An ultraviolet spectrophotometer was used for the detection of purity and concentration of total RNA, and agarose gel electrophoresis was used for the detection of RNA integrity. The concentration of RNA was adjusted to 500 ng/μl. A reverse transcription kit was used for the reverse transcription of total RNA into cDNA, in strict accordance with the manufacturer's instructions. The reaction conditions were 42˚C for 60 min, 95˚C for 3 min. SYBR Premix Ex Taq™ kit and quantitative PCR detector were used for PCR. The RT-qPCR system was: 20 μl total volume, 10.0 μl of SYBR Premix Ex Taq™ (2X), 0.4 μl of upstream primers and 0.4 μl of downstream primers (10 μM), 0.4 μl of ROX reference dye II, 2.0 μl of template, 6.8 μl of double distilled water (ddH_2O). qPCR reaction conditions were: Pre-denaturation at 95˚C for 10 min, 45 cycles of 95˚C for 10 sec, 60˚C for 30 sec, 72˚C for 10 sec; the procedure was repeated 3 times. The above system was configured following strictly the manufacturer's instructions. U6 was used as the internal reference of miR-378e and the U6 primers were synthesized by Shanghai Shenggong Biology Engineering Technology Service, Ltd.: Upstream, 5'-CTC GCT TCG GCA GCA CA-3' reverse transcription and downstream, 5'-AAC GCT TCA CGA ATT TGC GT-3'. The upstream and downstream primers of miR-378e were synthesized by Guangzhou Ruibo Co., Ltd.: Upstream, 5'-TTC GAG CCT ACT GGA CTT GGA G-3' and downstream, 5'-AGG GTC CGA GGT ATT CGC ACT-3' (Fig. 1). The $2^{-\Delta Cq}$ method (21) was used to quantify the relative expression levels of miR-378e in the blood.

Detection of LI-cadherin expression by ELISA. Operation steps: Blank well (the blank control well was the same as earlier steps; however, no enzyme labeling reagents and samples were added), standard well and the well of sample to be tested were respectively set. Standard sample (50 μl) was accurately added into the reaction well in which the enzyme label was coated. Firstly, 40 μl of the sample dilution were added into the well,

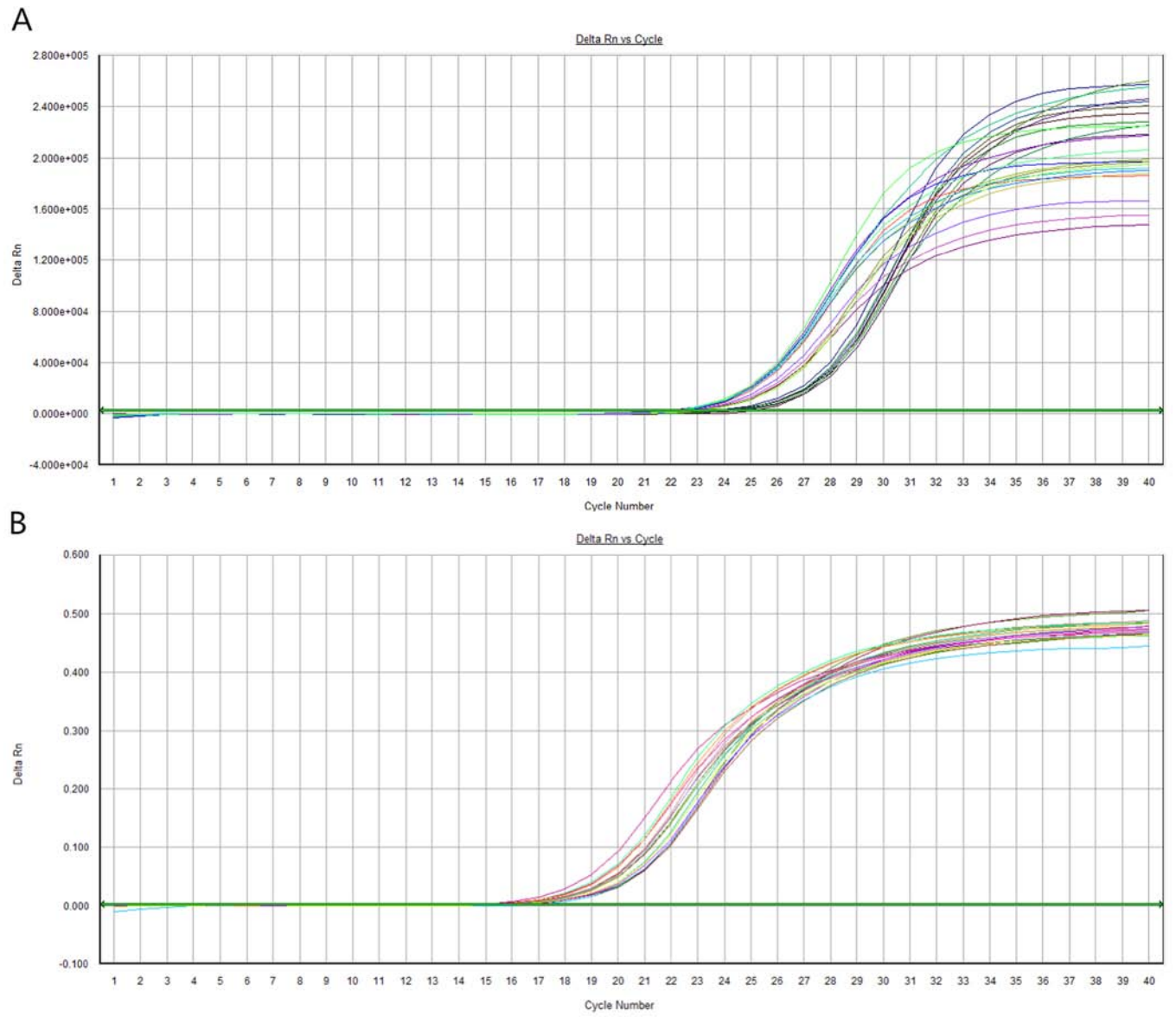

Figure 1. Amplification curves of (A) the target gene and (B) the reference gene.

and then, 10 μl of the sample to be tested were added into it (the final sample was diluted 5 times). The reaction well was sealed with a sealing film and incubated in a water bath or an incubator for 30 min. After the sealing film was removed, the liquid was discarded. The well was dried with absorbent paper and was filled with washing liquid. After standing for 30 sec, this step was repeated 5 times, and the reaction well was dried. In addition to the blank well, 50 μl of the enzyme labeling reagent were added into each well, and the mixture was incubated at 37˚C for 30 min. Next, the wells were washed. Substrate A (50 μl) and substrate B were added into each well, and the color was developed at 37˚C for 15 min in the dark. Stop solution (50 μl) was added into each well, and the absorbance (OD value) of each well was measured at a wavelength of 450 nm in 25 min using a BS-1101 enzyme microplate reader. The expression level of LI-cadherin was calculated.

Follow-up and observation indicators. Patients with colorectal cancer were followed up by hospital re-examination and telephone calls. The survival time of the patients was recorded at 1, 2 and 3 years after leaving hospital. The differences in the expression levels of LI-cadherin and miR-378e between the experimental and the control group were observed. The association of the expression levels of LI-cadherin and miR-378e with the clinical stage and the differentiation degree was analyzed according to the clinicopathological features of the patients with colorectal cancer. The value of the single and the combined detection of LI-cadherin and miR-378e, as well as their prognostic significance were analyzed.

Statistical analysis. SPSS 19.0 software (IBM Corp.) was used for the statistical analysis of the experimental data. Enumeration data were expressed as percentages (%) and Chi-square (χ^2) test was used for their comparison between groups. Measurement data were expressed as the mean ± SD and t-test was used for their comparison between two groups. ROC curve analysis was used to assess the sensitivity and specificity of the single and the combined detection. Kaplan-Meier

Table I. Comparison of the general data between the two groups [n (%), mean ± SD].

Characteristics	Experimental group (n=110)	Control group (n=90)	χ^2/t	P-value
Sex			0.101	0.751
Male	66 (60.00)	52 (57.78)		
Female	44 (40.00)	38 (42.22)		
Age (years)	62.13±9.21	61.89±9.28	0.183	0.855
Body mass index (kg/m^2)	19.79±3.21	19.83±3.57	0.083	0.934
Average height (cm)	167.34±4.43	166.62±4.64	1.119	0.264
History of smoking			0.758	0.384
Yes	63 (57.27)	57 (63.33)		
No	47 (42.73)	33 (36.67)		
Hypertension			0.419	0.517
Yes	39 (35.45)	28 (31.11)		
No	71 (64.55)	62 (68.89)		
Alcohol consumption			1.115	0.291
Yes	81 (73.64)	72 (80.00)		
No	29 (26.36)	18 (20.00)		
HB (gm/dl)	12.11±1.81	12.24±2.01	0.481	0.631
PLT (x10^9/l)	155.78±21.87	158.31±22.09	0.810	0.419
WBC (x10^9/l)	7.13±2.34	7.24±2.17	0.342	0.733
RBC (x10^{12}/l)	4.43±0.76	4.32±0.81	0.989	0.324
ALT (U/l)	23.14±10.32	22.89±10.12	0.172	0.864
AST (U/l)	19.38±7.69	19.67±6.65	0.282	0.778

HB, hemoglobin; PLT, platelet; WBC, white blood cells; RBC, red blood cells; ALT, alanine transaminase; AST, aspartate transaminase.

analysis and log-rank test were used for survival analysis. Pearson's correlation coefficient was used to analyze bivariate normal distribution data. P<0.05 was considered to indicate a statistically significant difference.

Results

Comparison of the general data between the two groups. The general clinical baseline data of the experimental and control group were collected, including sex, age, body mass index, average height, history of smoking, hypertension, alcohol consumption, hemoglobin (HB), platelet (PLT), white blood cells (WBC), red blood cells (RBC), alanine transaminase (ALT) and aspartate transaminase (AST). The results of their statistical analysis revealed that there was no statistically significant difference in the data between the two groups (P>0.05) (Table I).

Comparison of the expression levels of LI-cadherin and miR-378e in the serum of the two groups. The expression levels of LI-cadherin and miR-378e in the serum of all research subjects were detected. As shown in Table II, the expression level of LI-cadherin in the control group was higher than that in the experimental group, and the difference was statistically significant (P<0.001); the expression level of miR-378e in the control group was significantly higher than that in the experimental group (P<0.001).

Table II. Comparison of LI-cadherin and miR-378e serum expression levels between the two groups (mean ± SD).

Groups	LI-cadherin (ng/ml)	miR-378e
Experimental group (n=110)	4.11±1.57	4.53±1.88
Control group (n=90)	7.34±1.86	8.59±2.12
t	-10.88	15.63
P-value	<0.001	<0.001

Relationship between the LI-cadherin and miR-378e expression levels and the clinicopathological characteristics of the patients in the experimental group. The clinicopathological characteristics of 110 patients diagnosed with colorectal cancer were recorded. According to the results of the statistical analysis, LI-cadherin was not significantly associated with sex, age or histological type (P>0.05); however, LI-cadherin was significantly associated with the pathogenic site, the lymphatic metastasis, depth of infiltration, degree of differentiation and clinical stage (P<0.05). The expression level of miR-378e was not associated with sex, age, pathogenic site, lymphatic metastasis, degree of differentiation, clinical stage or histological type (P>0.05);

Table III. Relationship between the LI-cadherin and miR-378e expression levels and the clinicopathological characteristics of the patients in the experimental group (mean ± SD).

Characteristics	Cases	LI-cadherin (ng/ml))	t	P-value	miR-378e	t	P-value
Sex			0.136	0.892		0.304	0.761
Male	66	4.09±1.58			4.43±1.92		
Female	44	4.12±1.53			4.51±1.76		
Age (years)			0.665	0.507		0.345	0.730
≤50	34	3.97±1.55			4.45±1.87		
>50	76	4.12±1.63			4.54±1.79		
Site			2.851	0.005		1.102	0.272
Rectum	52	4.17±1.65			4.37±1.85		
Colon	58	3.46±1.87			4.65±1.71		
Lymphatic metastasis			2.341	0.020		0.488	0.626
Yes	45	3.47±1.74			4.38±1.68		
No	65	4.02±1.54			4.50±1.79		
Depth of infiltration			3.114	0.002		3.419	<0.001
T1+T2	42	4.11±1.21			4.01±1.57		
T3+T4	68	3.76±1.86			4.87±2.12		
Degree of differentiation			10.86	<0.001		0.348	0.728
High and medium	71	4.21±1.76			4.61±1.98		
Low	39	2.01±1.19			4.52±1.85		
Clinical stage			8.886	<0.001		0.281	0.779
A+B	69	4.10±1.65			4.47±1.99		
C+D	41	2.25±1.43			4.54±1.69		
Histological type			1.246	0.214		0.695	0.488
Tubular adenocarcinoma	58	3.69±1.78			4.51±1.53		
Non-tubular adenocarcinoma	52	3.98±1.67			4.67±1.87		

however, miR-378e was significantly associated with the depth of infiltration (P<0.05) (Table III).

Comparison of the value of single detection and combined detection of LI-cadherin and miR-378e in the diagnosis of colorectal cancer. As presented in Table IV, the sensitivity and specificity of LI-cadherin were 84 and 80%, respectively, and the optimal critical value was 5.76. The sensitivity and specificity of miR-378e were 89 and 80%, respectively, and the optimal critical value was 6.06. The sensitivity and specificity of the combined diagnosis were 86 and 94%, respectively. miR-378e detection had the highest sensitivity, and the combined detection had the highest specificity. The Youden indexes were 0.82, 0.68 and 0.64 for the combined detection, miR-378e, and LI-cadherin, respectively. The larger the Youden index was, the better the effect of detection was, and the higher the authenticity. ROC curves are shown in Figs. 2 and 3).

Survival of patients in the experimental group

Relationship between the expression of LI-cadherin and prognosis. The survival data of the experimental group were analyzed and the optimal threshold (5.76) of the expression level of LI-cadherin was taken as the limit. There were 88 cases in the low-expression group, in which the value of LI-cadherin was <5.76, and 22 cases in the high-expression group, in which the value of LI-cadherin was ≥5.76. The deadline of the follow-up period was November 20, 2017. The survival rate in the high-expression group was 63.64%, and the survival rate in the low-expression group was 39.77%. As shown in Fig. 4, the survival rate of patients in the LI-cadherin high-expression group was significantly higher than that in the low-expression group (P<0.05).

Relationship between the expression of miR-378e and prognosis. The survival data of the experimental group were analyzed, and the optimal threshold (6.06) of the expression level of miR-378e was taken as the limit. There were 90 cases in the low-expression group, in which the value of miR-378e was <6.06, and 20 cases in the high-expression group, in which the value of miR-378e was ≥6.06. The deadline of the follow-up period was November 20, 2017. The survival rate in the high-expression group was 65.00%, and the survival rate in the low-expression group was 40.00%. As shown in Fig. 5, the survival rate of patients in the miR-378e high-expression group was significantly higher than that in the low expression group (P<0.05).

Correlation analysis of LI-cadherin and miR-378e. The results of Pearson's correlation coefficient analysis revealed that there

Table IV. Comparison of the single detection and the combined detection of LI-cadherin and miR-378e in the diagnosis of colorectal cancer.

Items	Sensitivity (%)	Specificity (%)	Youden index	Optimal threshold
LI-cadherin	84	80	0.64	5.76
miR-378e	89	80	0.68	6.06
LI-cadherin + miR-378e	86	94	0.82	-

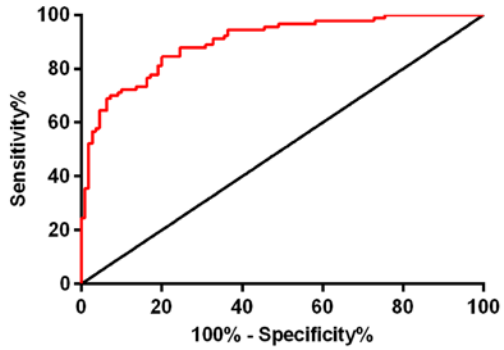

Figure 2. ROC curve of LI-cadherin single-color detection in colorectal cancer. The area under the ROC curve for LI-cadherin in serum was 0.8993 (95% CI, 0.8572-0.9414). The sensitivity and specificity were 84 and 80%, respectively, and the optimal threshold was 5.76.

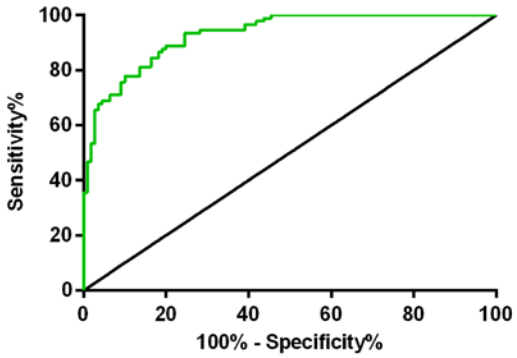

Figure 3. ROC curve of miR-378e single-color detection in colorectal cancer. The area under the ROC curve for miR-378e in serum was 0.9298 (95% CI, 0.8974-0.9622). The sensitivity and specificity were 89 and 80%, respectively, and the optimal threshold was 6.06.

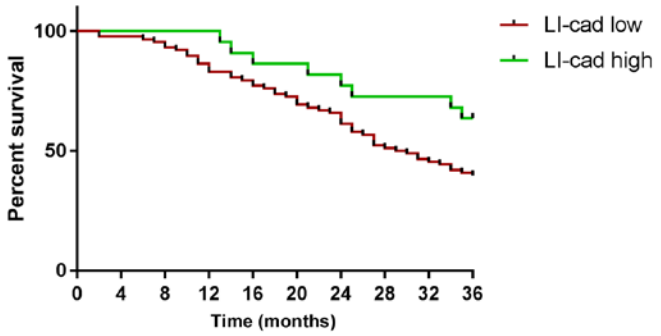

Figure 4. Relationship between LI-cadherin expression and survival. At the end of the follow-up period, the survival rate of the patients in the high-expression group was 63.64%, whereas in the low-expression group was 39.77%. The survival rate of patients with high expression of LI-cadherin was significantly higher than that of the patients with low expression (P<0.05).

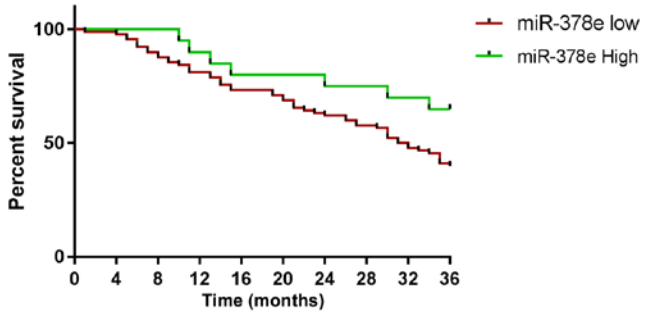

Figure 5. Relationship between miR-378e expression and survival. At the end of the follow-up period, the survival rate of the patients in the high-expression was 65.00%, whereas in the low-expression group was 40.00%. The survival rate of patients with high expression of miR-378e was significantly higher than that of the patients with low expression (P<0.05).

was a significantly positive correlation between LI-cadherin and miR-378e in the experimental, as well as the control group (r=0.5845 and 0.6356, respectively; P<0.05) (Figs. 6 and 7).

Discussion

Colorectal cancer is one of the most common malignant tumors with high morbidity, that ranks second among female malignant tumors and third among male malignant tumors worldwide (22). Colorectal cancer originates from the epithelial cells in colon or rectum of the digestive tract, which is also one of the main culprits that are responsible for the cancer-related deaths in humans (23,24). Therefore, the accurate diagnosis of colorectal cancer is crucial. In addition, the occurrence, growth, infiltration and metastasis of tumors are extremely complex processes, and genes and factors that affect their processes have become hotspots in the researche of cancer (1,25). Moreover, the therapeutic efficacy of colorectal cancer and the improvement of patients' prognosis mainly depend on early diagnosis and treatment (26).

LI-cadherin is a new member of the cadherin superfamily. The functional characteristics and the unique structure differentiate LI-cadherin from the classical cadherins (27). LI-cadherin regulates the adhesion function of cells through the complex structure of the cytoplasmic region, its diverse functions, and the interaction with calnexin (9). Relevant studies have shown that silencing LI-cadherin can increase the expression levels of MMP-2 and MMP-9 and downregulate the protein level of galectin-3, which is a substrate of MMP-2 and MMP-9 (28). Downregulation of the LI-cadherin expression

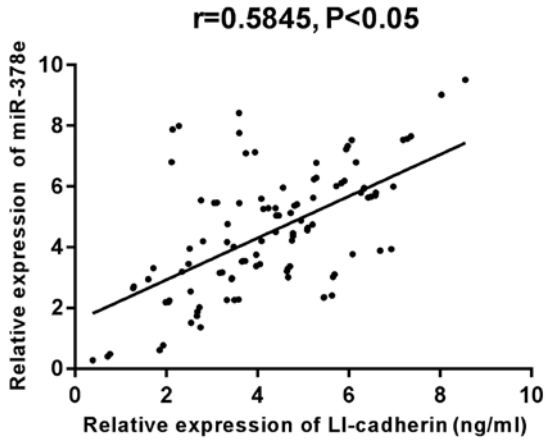

Figure 6. Correlation analysis of LI-cadherin and miR-378e expression levels in the experimental group. According to Pearson's correlation coefficient analysis, LI-cadherin was positively correlated with miR-378e in the experimental group (r=0.5845, P<0.05).

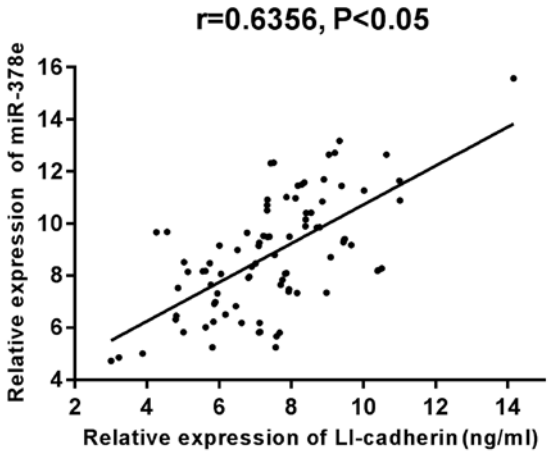

Figure 7. Correlation analysis of LI-cadherin and miR-378e expression levels in the control group. According to Pearson's correlation coefficient analysis, LI-cadherin was positively correlated with miR-378e in the control group (r=0.6356, P<0.05).

can promote the invasion of cancer cells by enhancing the expression of MMP-2 and MMP-9, activating and degrading the ingredients of extracellular matrix, which can also facilitate the adhesion and migration of cancer cells by altering the expression of galectin-3 (28). In recent years, miRNA has been a research hotspot in the field of molecular biology and genetics, and mature miRNA molecules have been shown to combine with Argonaute protein and other molecules, forming an RNA-induced silencing complex in cells. They can match and combine with the untranslated region of target mRNA 3' end completely or incompletely to induce the degradation of target mRNA or block its post-transcriptional translation (29), to involve in a series of biological processes, including cell proliferation, apoptosis, differentiation, metabolism, development and tumor metastasis (20).

In the present study, the clinicopathological characteristics of the research subjects in the experimental and the control group were compared, and the two groups were shown to be comparable. Through immunoblotting experiment analysis, Bernhard *et al* (30) found that LI-cadherin may be a potential biomarker when colon cancer cells secrete into plasma. In the present study, by detecting the expression levels of LI-cadherin and miR-378e in the serum, it was shown that the expression levels of LI-cadherin and miR-378e in the control group were significantly higher than those in the experimental group (P<0.001), which indicates that LI-cadherin and miR-378e are expressed at low levels in colorectal cancer. When the expression of LI-cadherin is reduced in patients with colon cancer, tumors tend to be poorly differentiated and have the characteristics of proliferation, invasion and metastasis (13). However, Gao *et al* (20) have reported that miR-378e is significantly downregulated in colorectal cancer tissues, suggesting that miR-378e may play a similar role in cancer inhibition in the occurrence and development of tumors. Cadherin not only plays an important role in the intercellular adhesion of epithelial cells, but also maintains the intact form of the epithelial cell lines of tumors. Once the intact form of the epithelial cell lines is lost, cancer cell lines will have the ability to invade (31). The expression of LI-cadherin is correlated with the dedifferentiation level of tumors, lymph node metastasis, and TNM stage and progression of tumors. The prognosis of patients with tumors is closely related to these factors (31). Our results revealed that LI-cadherin was significantly associated with the pathogenic site, the lymphatic metastasis, depth of infiltration, degree of differentiation, and clinical stage (P<0.05). This indicates that cancer cells whose differentiation degree is good can maintain the good expression ability of LI-cadherin, while those with poor differentiation degree have lower expression ability. The expression level of LI-cadherin is related to lymph node metastasis, indicating that LI-cadherin can be used as an indicator to estimate the prognosis of patients with colorectal cancer, and has some value in predicting the survival time of patients with colorectal cancer and the occurrence and development of this disease. However, miR-378e is only related to the depth of infiltration, which suggests that the infiltration of colorectal cancer may induce the expression of miR-378e, indicating that miR-378e can be considered as a tumor suppressor gene. Up to our knowledge, there are still few relevant studies on the specific mechanisms of miR-378e, thus, further in-depth research is needed. According to the results of the present study, miR-378e had the highest sensitivity, and the combined detection had the highest specificity. The Youden index was the highest for the combined detection, followed by that of the miR-378e detection, and that of LI-cadherin single detection. Therefore, the combined detection is more valuable than the single detection in the diagnosis of colorectal cancer. In order to further investigate the correlation of LI-cadherin expression level with the miR-378e expression level in colorectal cancer, a series of animal experiments and clinical experiments need to be carried out. The analysis of the survival of the patients in the experimental group, revealed that the high expression of LI-cadherin is associated to the high survival rate of the patients, and the low LI-cadherin expression is associated to a lower survival rate, suggesting that LI-cadherin can be used as a biomarker to predict the prognosis of patients with colorectal cancer, which is similar to the research results of Takamura *et al* (32). There are few studies on the relationship betweeen miR-378e and tumor prognosis. Donnarumma *et al* (33) have reported that patients with breast cancer, whose miR-378 level is low, have a long overall survival time, suggesting that miR-378e has an effect on the prognosis

of the disease (P<0.01). The results of the present study demonstrated that patients with high expression of miR-378e have high survival rate, and patients with low miR-378e expression have a lower survival rate, indicating that miR-378e can also be used as a biomarker for the prognosis of patients with colorectal cancer. Different miRNAs have different expression levels in tumors, thus, more studies in this direction need to be carried out. In the present study, there was a significantly positive correlation between LI-cadherin and miR-378e in both groups, indicating that the changes in the expression of LI-cadherin may be related to miR-378e. Currently, there is little research on the correlation between LI-cadherin and miR-378e, therefore future studies need to be conducted to verity this result.

In the present study the expression levels of LI-cadherin and miR-378e, as well as and their prognostic value in colorectal cancer, were investigated in a comprehensive way to provide future reference for clinical researches. However, the specific mechanisms of LI-cadherin and miR-378e and their effects on different cancers need to be further investigated. The relationship between clinicopathological factors and prognosis need to be analyzed multifactorially to estimate the prognosis of patients more accurately.

In conclusion, LI-cadherin and miR-378e expression levels may contribute to the understanding of the occurrence, development and biological behavior of colorectal cancer. LI-cadherin and miR-378e expression levels have positive diagnostic value for colorectal cancer and can be used as biomarkers for the prognosis of colorectal cancer.

Acknowledgements

Not applicable.

Funding

No funding was received.

Availability of data and materials

The datasets used and/or analyzed during the present study are available from the corresponding author on reasonable request.

Authors' contributions

XW interpreted the data and drafted the manuscript. XW and ZHL performed PCR. JF and WX collected and analyzed the general data. XW and ZXL were responsible for ELISA. All authors read and approved the final manuscript.

Ethics approval and consent to participate

The study was approved by the Ethics Committee of Weihai Central Hospital (Weihai, China). All participants had complete clinical data. Signed written informed consents were obtained from the participants of this study and/or their guardians.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Ferlay J, Shin HR, Bray F, Forman D, Mathers C and Parkin DM: Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. Int J Cancer 127: 2893-2917, 2010.
2. Kwon CH, Park HJ, Choi JH, Lee JR, Kim HK, Jo HJ, Kim HS, Oh N, Song GA and Park DY: Snail and serpinA1 promote tumor progression and predict prognosis in colorectal cancer. Oncotarget 6: 20312-20326, 2015.
3. Koerner J, Brunner T and Groettrup M: Inhibition and deficiency of the immunoproteasome subunit LMP7 suppress the development and progression of colorectal carcinoma in mice. Oncotarget 8: 50873-50888, 2017.
4. Sun X, Wang T, Zhang C, Ning K, Guan ZR, Chen SX, Hong TT and Hua D: S100A16 is a prognostic marker for colorectal cancer. J Surg Oncol 117: 275-283, 2018.
5. Edwards BK, Ward E, Kohler BA, Eheman C, Zauber AG, Anderson RN, Jemal A, Schymura MJ, Lansdorp-Vogelaar I, Seeff LC, *et al*: Annual report to the nation on the status of cancer, 1975-2006, featuring colorectal cancer trends and impact of interventions (risk factors, screening, and treatment) to reduce future rates. Cancer 116: 544-573, 2010.
6. Bagaria B, Sood S, Sharma R and Lalwani S: Comparative study of CEA and CA19-9 in esophageal, gastric and colon cancers individually and in combination (ROC curve analysis). Cancer Biol Med 10: 148-157, 2013.
7. Tan E, Gouvas N, Nicholls RJ, Ziprin P, Xynos E and Tekkis PP: Diagnostic precision of carcinoembryonic antigen in the detection of recurrence of colorectal cancer. Surg Oncol 18: 15-24, 2009.
8. Flamini E, Mercatali L, Nanni O, Calistri D, Nunziatini R, Zoli W, Rosetti P, Gardini N, Lattuneddu A, Verdecchia GM, *et al*: Free DNA and carcinoembryonic antigen serum levels: An important combination for diagnosis of colorectal cancer. Clin Cancer Res 12: 6985-6988, 2006.
9. Bartolmäs T, Hirschfeld-Ihlow C, Jonas S, Schaefer M and Gessner R: LI-cadherin cis-dimerizes in the plasma membrane Ca (2+) independently and forms highly dynamic trans-contacts. Cell Mol Life Sci 69: 3851-3862, 2012.
10. Gessner R and Tauber R: Intestinal cell adhesion molecules. Liver-intestine cadherin. Ann N Y Acad Sci 915: 136-143, 2000.
11. Ordóñez NG: Cadherin 17 is a novel diagnostic marker for adenocarcinomas of the digestive system. Adv Anat Pathol 21: 131-137, 2014.
12. Snow AN, Mangray S, Lu S, Clubwala R, Li J, Resnick MB and Yakirevich E: Expression of cadherin 17 in well-differentiated neuroendocrine tumours. Histopathology 66: 1010-1021, 2015.
13. Wang Y, Gao X, Wei F, Zhang X, Yu J, Zhao H, Sun Q, Yan F, Yan C, Li H, *et al*: The hOGG1 Ser326Cys polymorphism contributes to digestive system cancer susceptibility: Evidence from 48 case-control studies. Tumour Biol 36: 1029-1038, 2015.
14. Farazi TA, Hoell JI, Morozov P and Tuschl T: MicroRNAs in human cancer. Adv Exp Med Biol 774: 1-20, 2013.
15. Romero-Cordoba SL, Salido-Guadarrama I, Rodriguez-Dorantes M and Hidalgo-Miranda A: miRNA biogenesis: Biological impact in the development of cancer. Cancer Biol Ther 15: 1444-1455, 2014.
16. Jiang S, Zhang HW, Lu MH, He XH, Li Y, Gu H, Liu MF and Wang ED: MicroRNA-155 functions as an OncomiR in breast cancer by targeting the suppressor of cytokine signaling 1 gene. Cancer Res 70: 3119-3127, 2010.
17. Lee EJ, Gusev Y, Jiang J, Nuovo GJ, Lerner MR, Frankel WL, Morgan DL, Postier RG, Brackett DJ and Schmittgen TD: Expression profiling identifies microRNA signature in pancreatic cancer. Int J Cancer 120: 1046-1054, 2007.
18. Magrelli A, Azzalin G, Salvatore M, Viganotti M, Tosto F, Colombo T, Devito R, Di Masi A, Antoccia A, Lorenzetti S, *et al*: Altered microRNA expression patterns in hepatoblastoma patients. Transl Oncol 2: 157-163, 2009.
19. Lu J, Getz G, Miska EA, Alvarez-Saavedra E, Lamb J, Peck D, Sweet-Cordero A, Ebert BL, Mak RH, Ferrando AA, *et al*: MicroRNA expression profiles classify human cancers. Nature 435: 834-838, 2005.
20. Gao L, Tian Y, Zhao Z, Zhang L, He X and Pei Y: Expression and clinical significance of miR-224 and miR-378e in colorectal cancer tissues. Tianjin Med J 8: 737-739, 20113.

21. Tang R, Liang L, Luo D, Feng Z, Huang Q, He R, Gan T, Yang L and Chen G: Downregulation of miR-30a is associated with poor prognosis in lung cancer. Med Sci Monit 21: 2514-2520, 2015.
22. Torre LA, Bray F, Siegel RL, Ferlay J, Lortet-Tieulent J and Jemal A: Global cancer statistics, 2012. CA Cancer J Clin 65: 87-108, 2015.
23. Alnabulsi A and Murray GI: Integrative analysis of the colorectal cancer proteome: Potential clinical impact. Expert Rev Proteomics 13: 917-927, 2016.
24. Chen Z, Gao S, Wang D, Song D and Feng Y: Colorectal cancer cells are resistant to anti-EGFR monoclonal antibody through adapted autophagy. Am J Transl Res 8: 1190-1196, 2016.
25. Dimofte G, Târcoveanu E, Taraşi M, Panait C, Lozneanu G, Nicolescu S, Porumb V and Grigoraş O: Mean number of lymph nodes in colonic cancer specimen: Possible quality control index for surgical performance. Chirurgia (Bucur) 106: 759-764, 2011.
26. Stojkovic Lalosevic M, Stankovic S, Stojkovic M, Markovic V, Dimitrijevic I, Lalosevic J, Petrovic J, Brankovic M, Pavlovic Markovic A and Krivokapic Z: Can preoperative CEA and CA19-9 serum concentrations suggest metastatic disease in colorectal cancer patients? Hell J Nucl Med 20: 41-45, 2017.
27. Li R, Yang HQ, Xi HL, Feng S and Qin RH: Inhibition of CDH17 gene expression via RNA interference reduces proliferation and apoptosis of human MKN28 gastric cancer cells. Int J Oncol 50: 15-22, 2017.
28. Yu Q, Shen W, Zhou H, Dong W and Gao D: Knockdown of LI-cadherin alters expression of matrix metalloproteinase-2 and -9 and galectin-3. Mol Med Rep 13: 4469-4474, 2016.
29. Bartel DP: MicroRNAs: Genomics, biogenesis, mechanism, and function. Cell 116: 281-297, 2004.
30. Bernhard OK, Greening DW, Barnes TW, Ji H and Simpson RJ: Detection of cadherin-17 in human colon cancer LIM1215 cell secretome and tumour xenograft-derived interstitial fluid and plasma. Biochim Biophys Acta 1834: 2372-2379, 2013.
31. van der Wurff AA, Vermeulen SJ, van der Linden EP, Mareel MM, Bosman FT and Arends JW: Patterns of alpha- and beta-catenin and E-cadherin expression in colorectal adenomas and carcinomas. J Pathol 182: 325-330, 1997.
32. Takamura M, Ichida T, Matsuda Y, Kobayashi M, Yamagiwa S, Genda T, Shioji K, Hashimoto S, Nomoto M, Hatakeyama K, *et al*: Reduced expression of liver-intestine cadherin is associated with progression and lymph node metastasis of human colorectal carcinoma. Cancer Lett 212: 253-259, 2004.
33. Donnarumma E, Fiore D, Nappa M, Roscigno G, Adamo A, Iaboni M, Russo V, Affinito A, Puoti I, Quintavalle C, *et al*: Cancer-associated fibroblasts release exosomal microRNAs that dictate an aggressive phenotype in breast cancer. Oncotarget 8: 19592-19608, 2017.