European Review for Medical and Pharmacological Sciences 2020; 24: 8801-8811

Hsa-miR-4730 as a new and potential diagnostic and prognostic indicators for pancreatic cancer

W. YANG[1], H.-Y. JU[2], X.-F. TIAN[3]

[1]Department of Thyroid and Breast Surgery, China-Japan Union Hospital of Jilin University, Changchun, Jilin Province, China
[2]Jilin Province Blood Center (Changchun Center Blood Station), Changchun, Jilin Province, China
[3]Department of Hepatobiliary and Pancreatic Surgery, China-Japan Union Hospital of Jilin University, Changchun, Jilin Province, China

Abstract. – OBJECTIVE: Pancreatic cancer is a gastrointestinal tumor with the highest malignancy and few diagnostic and prognostic markers. Patients with disease have a 5-year survival rate that is not more than 10%. As a research hotspot in recent years, miRs (microRNAs) are differentially expressed in various tumors, so they can be used as the potential diagnostic and prognostic markers. In this study, differentially expressed miRs in patients with pancreatic cancer were screened out through the GEO chip, to provide potential markers for clinical practice. This study aimed to explore the expression and potential value of miR-4730 in pancreatic cancer.

PATIENTS AND METHODS: Differentially expressed miRs in pancreatic cancer were analyzed through logging in GEO DataSets to download GSE112264. Fifty patients with pancreatic cancer who were treated in our hospital from May 2012 to January 2014 (Group A), 50 patients with benign pancreatic lesions during the same period (Group B), and 50 healthy individuals undergoing physical examinations (Group C) were enrolled in this study. The expression of miR-4730 in the serum and the cancer tissue was detected by qRT-PCR. The correlation of miR-4730 with pathological data, and the diagnostic values of differential indicators in the data were analyzed. The patients were followed up for 5 years to observe the relationship between miR-4730 and their survival.

RESULTS: The analysis of the GEO chip showed 305 differentially expressed miRs, among which 225 were highly expressed and 80 were lowly expressed, with miR-4730 differentially expressed most. The expression of serum miR-4730 in Group A was significantly lower than that in Groups B and C ($p<0.05$), so miR-4730 had a diagnostic value. The expression of miR-4730 in the cancer tissue was significantly lower than that in the adjacent tissue. The correlation analysis showed that the expression of miR-4730 in the cancer tissue was positively correlated with that in the serum. Patients with low miR-4730 expression had poorly differentiated pancreatic cancer, and patients with stages III+IV of pancreatic cancer had higher incidences of lymphatic invasion and distal metastasis ($p<0.05$), so miR-4730 had a diagnostic value. The 3- and 5-year survival rates in the high miR-4730 expression group were higher than those in the low expression group (both $p<0.05$). TNM staging, lymphatic invasion, distal metastasis, and miR-4730 were independent prognostic factors for the 3- and 5-year survival of patients with pancreatic cancer.

CONCLUSIONS: For patients with pancreatic cancer, those with low miR-4730 expression have poor survival and prognoses, so miR-4730 can be used as a potential observational index for the prognosis and diagnosis of the disease.

Key Words:
MiR-4730, Pancreatic cancer, Prognosis, GEO, Diagnosis, 5-year survival.

Introduction

Pancreatic cancer is the fourth leading cause of cancer-related death and the world's deadliest solid tumor, with a 5-year survival rate of 5% and median survival time less than 6 months. Although the survival rate has recently increased, it is still not more than 10%[1-3]. The disease is only cured by surgery at present, but few patients with stage I pancreatic cancer can be clinically diagnosed, because 80% of patients with pancreatic cancer have been in the advanced stage when diagnosed and cannot be operated[4]. Therefore,

Corresponding Author: Xiaofeng Tian, MD; e-mail: jlccyangwei@jlu.edu.cn

it is essential for the early diagnosis, which is considered as the preferred plan to improve the patients' survival, to find out highly specific and sensitive diagnostic indicators.

In addition to pathological biopsy, magnetic resonance imaging/magnetic resonance cholangiopancreatography (MRI/MRCP) are the most accurate methods for the diagnosis of pancreatic cancer, but they are expensive[5]. Relatively common blood test causes less trauma to patients than pathological biopsy and is cheaper than MRI and MRCP[6]. Therefore, a molecular biomarker for diagnosing the disease should be found out to improve the current situation. Currently, CA199 is the marker with the highest sensitivity for the diagnosis. It is differentially expressed in acute and chronic pancreatitis and cholecystitis, biliary obstruction, and hepatitis, and lowly expressed in poorly differentiated tumors[7].

As a non-coding short-strand RNA with a length of about 22-25 nt, a microRNA (miR) is expressed in many eukaryotes and viruses[8]. It negatively regulates gene expressions by binding to partial and complementary sequences in the 3'-untranslated region (3'-UTR) of specific target mRNA, thereby degrading and inhibiting the target gene[9,10]. According to the latest studies, miRs are differentially expressed in various tumors[11], and they inhibit the development and progression of the tumors by directly targeting target genes[12]. In this study, differentially expressed miRs in pancreatic cancer were screened out through GEO (GSE112264) chip. The results showed that miR-4730 was significantly and differentially expressed in the cancer and adjacent tissues. Therefore, the clinical value of miR-4730 was further explored.

Patients and Methods

Collection of GEO Data

The National Center for Biotechnology Information (https://www.ncbi.nlm.nih.gov/, NCBI) was logged in to change All Databases in the top left corner to GEO DataSets. Then, keywords (pancreatic cancer and microRNA) were entered in the Search Bar, and Search was clicked. After rough searching, GSE112264 chip related to pancreatic cancer was selected, and its basic information is shown in Table I.

Screening of Differentially Expressed Genes From GSE112264

The website (https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE112264) was visited to download a matrix file (Series Matrix File), an expression file, and a platform file (GPL21263). A Perl script was used to group cancer patients (n=50) and non-cancer patients (n=41). The edgeR package in R was used to analyze differentially expressed miRs, with logFoldChange=2 and $p<0.0001$ set. A volcano map and a heat map were plotted.

Collection of Clinical Data

A total of 50 patients with pancreatic cancer who were treated in our hospital from May 2012 to January 2014 (Group A), 50 patients with benign pancreatic lesions during the same period (Group B), and 50 healthy individuals undergoing physical examinations (Group C) were enrolled in this study. Group A consisted of 32 males and 18 females, with an average age of 55.1±4.1 years. Group B consisted of 28 males and 22 females, with an average age of 54.2±3.7 years. Group C

Table I. Basic information of GSE112264.

Data	GSE112264
Time	
Submission date	Mar 23, 2018
Last update date	Mar 01, 2019
Contact name	Junpei Kawauchi
Address	
Organization name	Toray Industries, Inc.
Department	New Frontiers Research Laboratories
Street address	6-10-1
City	Kamakura
Country	Japan
ZIP/Postal code	248-0036
Organism	Homo sapiens
Experiment type	Non-coding RNA profiling by array
Platforms	GPL21263 3D-Gene Human miRNA V21_1.0.0

consisted of 30 males and 20 females, with an average age of 55.1±3.9 years. This investigation was approved by the Medical Ethics Committee of our hospital. The healthy individuals in Group C had normal laboratory and imaging tests. The inclusion criteria for patients were as follows: patients confirmed with pancreatic cancer by pathology and imaging; patients who met the 8th edition of TNM staging issued by the American Joint Committee on Cancer (AJCC) in 2017[13]; patients who were informed and who signed the informed consent form. The exclusion criteria for patients were as follows: patients who had received corresponding anti-tumor treatment before this study; patients with expected survival time <1 month; patients with incomplete clinical data; patients who did not cooperate in follow-up.

Sample Collection

Peripheral blood (5 mL) was respectively drawn from the research objects in Groups A, B and C, allowed to stand for 30 min, and then centrifuged at 3000 rpm for 10 min, to collect the supernatant for subsequent experiments. The cancer tissue and adjacent tissue samples were collected from the patients in Group A. The tissues were collected during surgery from those who could be operated, and during pathological biopsy from those who could not be operated.

Major Reagents and Instruments

A TRIzol reagent and a mirVana™ qRT-PCR miRNA detection kit (Invitrogen, Carlsbad, CA, USA, 15596018, AM1558). A 7500 PCR instrument and a TaqMan™ MicroRNA reverse transcription kit (Applied Biosystems, Foster City, CA, USA, 7500, 4366596).

Detection of MiR Expression

The TRIzol reagent was used to extract total RNA from the collected serum and tissues. An UV spectrophotometer (Thermo Fisher Scientific™, Waltham, MA, USA, 840-300400) and agarose gel electrophoresis were used to detect its purity, concentration, and integrity. The TaqMan™ MicroRNA reverse transcription kit (Applied Biosystems, Foster City, CA USA, 4366596) was used to reversely transcribe the total RNA, with the steps carried out in strict accordance with the manufacturer's instructions. After cDNA was collected, the mirVana™ qRT-PCR miRNA detection kit (Invitrogen, Carlsbad, CA, USA, AM1558) and the ABI 7500 (Applied Biosystems, Foster City, CA, USA, 7500, 4366596) were used for amplification. The system was as follows: 5 μL of mirVana 5 X PCR Buffer, 0.5 μL of 50 X ROX™, 1 μL of cDNA, each 0.5 μL of upstream and downstream primers, and Nuclease-free Water was finally added to make up to 20 μL. The conditions were as follows: pre-denaturation at 95°C for 3 min, denaturation at 95°C for 15 s, and annealing and extension at 60°C for 30 s, for a total of 40 cycles. Each sample was provided with 3 same wells, and the experiment was repeatedly conducted for 3 times. U6 was used as an internal reference and $2^{-\Delta\Delta ct}$ was used to analyze the data.

Follow-Up

The patients were followed up by telephone, out-patient services, and the review of case data for 5 years, to record their survival. The follow-up was carried out once every 3 months at the first year, and once every 4 months at the 4 following years.

Statistical Analysis

SPSS 20.0 (IBM, Armonk, NY, USA) was used for statistical analysis. GraphPad 7 (GraphPad Software, San Diego, CA, USA) was used to plot figures. Kolmogorov-Smirnov (K-S) test was used to analyze the distribution of measurement data. The data that conformed to normal distribution were expressed by mean ± standard deviation (Meas±SD). The comparison of the data between groups was analyzed by independent samples *t*-test and represented by *t*. Ranked data were analyzed by rank sum test and represented by Z. Count data were analyzed by chi-square test. Receiver operating characteristic (ROC) curves were plotted to show the diagnostic value. Kaplan-Meier (K-M) survival curves were plotted to show the patients' 5-year survival, and Log-rank test was used for its analysis. Multivariate Cox regression analysis was performed on the independent risk factors affecting the patients' prognoses. Comparison between multiple groups was analyzed by one-way analysis of variance and represented by F. $p<0.05$ indicated a statistically significant difference.

Results

Screening Results of Differentially Expressed Genes

The analysis of the GEO chip showed 305 differentially expressed miRs, among which 225 were highly expressed and 80 were lowly ex-

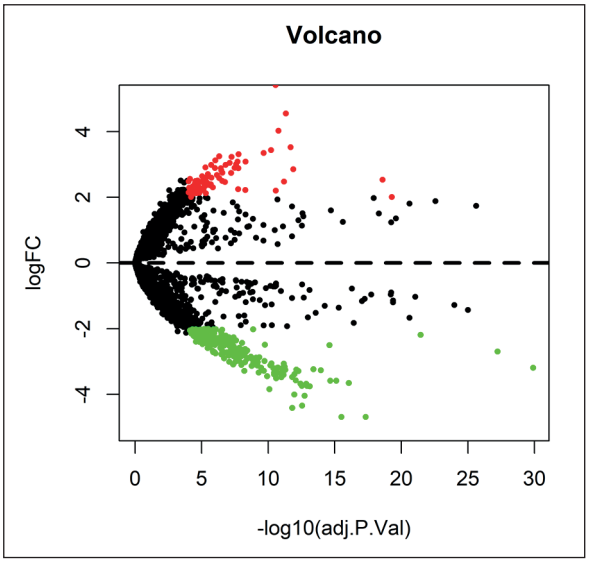

Figure 1. A volcano map for differentially expressed miRs. Red indicates high expression, green indicates low expression, and black indicates no difference.

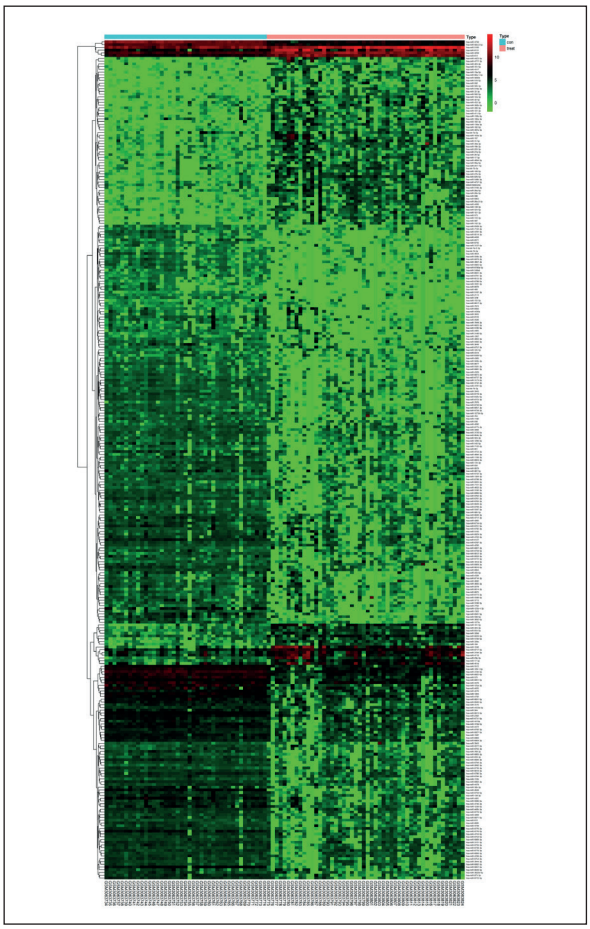

Figure 2. A heat map for differentially expressed miRs. Red indicates high expression and green indicates low expression (con indicates non-cancer patients and treat indicates cancer patients).

pressed, with miR-4730 differentially expressed most. Therefore, miR-4730 was chosen for this clinical study (Figures 1 and 2).

Expression and Diagnostic Value of MiR-4730

According to the detection, miR-4730 was differentially expressed in Groups A, B, and C ($p<0.001$). The expression of serum miR-4730 in Group A was significantly lower than that in Groups B and C ($p<0.05$), and the expression in Group B was significantly lower than that in Group C ($p<0.05$). According to the ROC curves, miR-4730 had a better diagnostic value for pancreatic cancer and benign pancreatic lesions (Table II). The expression of miR-4730 in the cancer tissue was significantly lower than that in the adjacent tissue

Table II. ROC parameters.

Data	GSE112264
Time	
Submission date	Mar 23, 2018
Last update date	Mar 01, 2019
Contact name	Junpei Kawauchi
Address	
Organization name	Toray Industries, Inc.
Department	New Frontiers Research Laboratories
Street address	6-10-1
City	Kamakura
Country	Japan
ZIP/Postal code	248-0036
Organism	Homo sapiens
Experiment type	Non-coding RNA profiling by array
Platforms	GPL21263 3D-Gene Human miRNA V21_1.0.0

(p<0.05). According to the correlation analysis, the expression in the cancer tissues was positively correlated with that in the serum (r=0.790, p<0.001; Figure 3).

Relationship Between MiR-4730 and Pathological Data

According to the median expression (0.769) of miR-4730, the patients in Group A were divided into high and low miR-4730 expression groups. Differences in the pathological data between the two groups were analyzed. The results showed that patients with low expression had poorly differentiated pancreatic cancer, and patients with stages III+IV of pancreatic cancer had higher incidences of lymphatic invasion and distal metastasis (p<0.05). The ROC curves were plotted according to the indicators with differences. The results showed that miR-4730 had a diagnostic value for differentiation, TNM staging, lymphatic invasion, and distal metastasis. See Tables III, IV and Figure 4.

Relationship Between MiR-4730 and Survival

All patients in Group A were followed up for 5 years, and their 3- and 5-year survival rates were 38.00% and 12.00%, respectively. The survival rates in the high miR-4730 expression group were significantly higher than those in the low expression group ($p_{3\text{-year}}$=0.002, $p_{5\text{-year}}$=0.004).

Cox Regression Analysis of 3- and 5-Year Survival

The Cox regression analysis was performed on the 3- and 5-year survival in Group A, and independent factors affecting the patients' prognoses were observed. The results showed that TNM staging (HR: 13.443, 95 CI%: 4.163-43.406), lymphatic invasion (HR: 0.121, 95 CI%: 0.029-0.500), distal metastasis (HR: 0.077, 95 CI%: 0.024-0.247), and miR-4730 (HR: 3.991, 95 CI%: 1.342-11.867) were independent prognostic factors affecting the patients' 3-year survival. Further analysis showed that TNM staging (HR: 8.065, 95 CI%: 3.353-19.394), lymphatic invasion (HR:

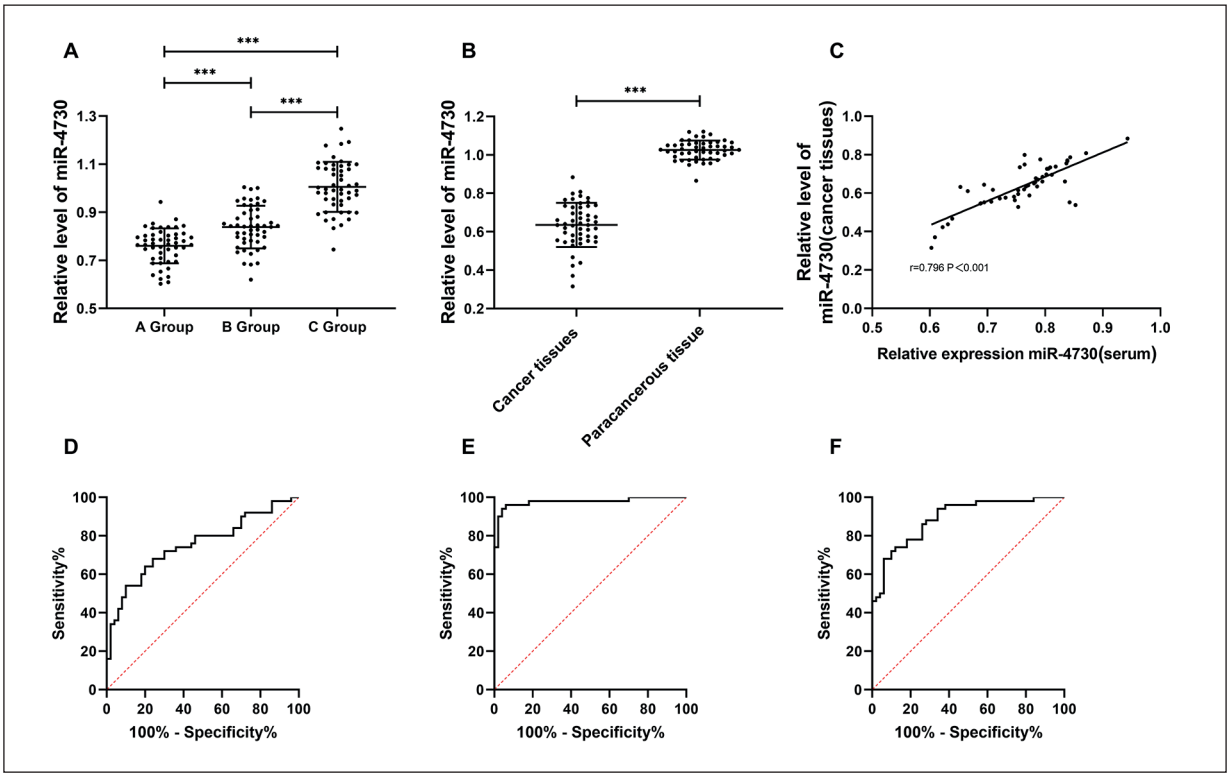

Figure 3. Diagnostic value and expression of miR-4730 in pancreatic cancer. **A**, miR-4730 expression in Groups A, B, and C; **B**, miR-4730 expression in cancer and adjacent tissues; **C**, The expression of miR-4730 in the cancer tissue was positively correlated with that in the serum of patients with pancreatic cancer; **D**, The diagnostic value of serum miR-4730 in Groups A and B; **E**, The diagnostic value of miR-4730 in Groups A and C; **F**, The diagnostic value of miR-4730 in Groups B and C. *** indicates p<0.001.

Table III. Relationship between miR-4730 and pathological data.

Parameters		Expression of miR-4730		χ^2-value	*p*-value
		Low expression	High expression		
Gender				0.347	0.556
	Male (n=32)	17 (68.00)	15 (60.00)		
	Female (n=18)	8 (32.00)	10 (40.00)		
Age				0.081	0.777
	≥ 55 years old (n=27)	14 (56.00)	13 (52.00)		
	< 55 years old (n=23)	11 (44.00)	12 (48.00)		
Lesion site				0.333	0.564
	Head of pancreas (n=30)	14 (56.00)	16 (64.00)		
	Others (n=20)	11 (44.00)	9 (36.00)		
Differentiation				6.650	0.010
	Poorly differentiated) (n=29	19 (76.00)	10 (40.00)		
	Moderately + highly differentiated (n=21)	6 (24.00)	15 (60.00)		
TNM staging				9.742	0.002
	Stages I+II (n=23)	6 (24.00)	17 (68.00)		
	Stages III+IV (n=27)	19 (76.00)	8 (32.00)		
Lymphatic invasion				8.333	0.004
	Yes (n=30)	20 (80.00)	10 (40.00)		
	No (n=20)	5 (20.00)	15 (60.00)		
Vascular invasion				0.325	0.569
	Yes (n=28)	15 (60.00)	13 (52.00)		
	No (n=22)	10 (40.00)	12 (48.00)		
Distant metastasis				15.909	< 0.001
	Yes (n=22)	18 (72.00)	4 (16.00)		
	No (n=28)	7 (28.00)	21 (84.00)		

0.240, 95 CI%: 0.098-0.590), distal metastasis (HR: 0.098, 95 CI%: 0.036-0.271), and miR-4730 (HR: 3.192, 95 CI%: 1.283-7.945) were independent prognostic factors affecting the patients' 5-year survival (Figure 5 and Table V).

Discussion

At present, pancreatic cancer is one of the malignant tumors with the highest mortality[14]. By monitoring the survival trend of global cancer patients from 2000 to 2014, Allemani et al[15] found that the 5-year net survival rate of patients with pancreatic cancer, the lowest one among all cancer patients, decreased from 14.4% in 2000 to 9.9% in 2014[15]. Early surgical treatment is the preferred plan to improve the patients' prognoses and survival, but there are few patients with stage I pancreatic cancer[16]. This may be because early pancreatic cancer is mainly manifested as abdominal pain and has no other clinical symptom. Additionally, the early diagnostic indicators for the disease are rare. Accordingly, there are few patients with stage I pancreatic cancer.

As an important serological marker, CA199 has high sensitivity but low specificity for the diagnosis of pancreatic cancer[17]. Abue et al[18] has shown that miRs, a research hotspot in recent years, are involved in the development and progression of pancreatic cancer. According to Li et al[19], the down-regulation of miR-506 regulates SPHK1/

Table IV. ROC parameters.

Factors	AUC	Std	95% CI	*p*-value	Specificity	Sensitivity	Youden index	Cut-off
Differentiation	0.800	0.063	0.677-0.922	< 0.001	58.62%	85.71%	44.33	> 0.755
TNM staging	0.821	0.059	0.706-0.937	< 0.001	100.00	59.24%	59.26	< 0.745
Lymphatic invasion	0.725	0.077	0.575-0.875	0.001	66.67%	80.00%	46.67%	> 0.766
Distant metastasis	0.825	0.059	0.709-0.941	< 0.001	90.90%	67.86%	58.77%	> 0.783

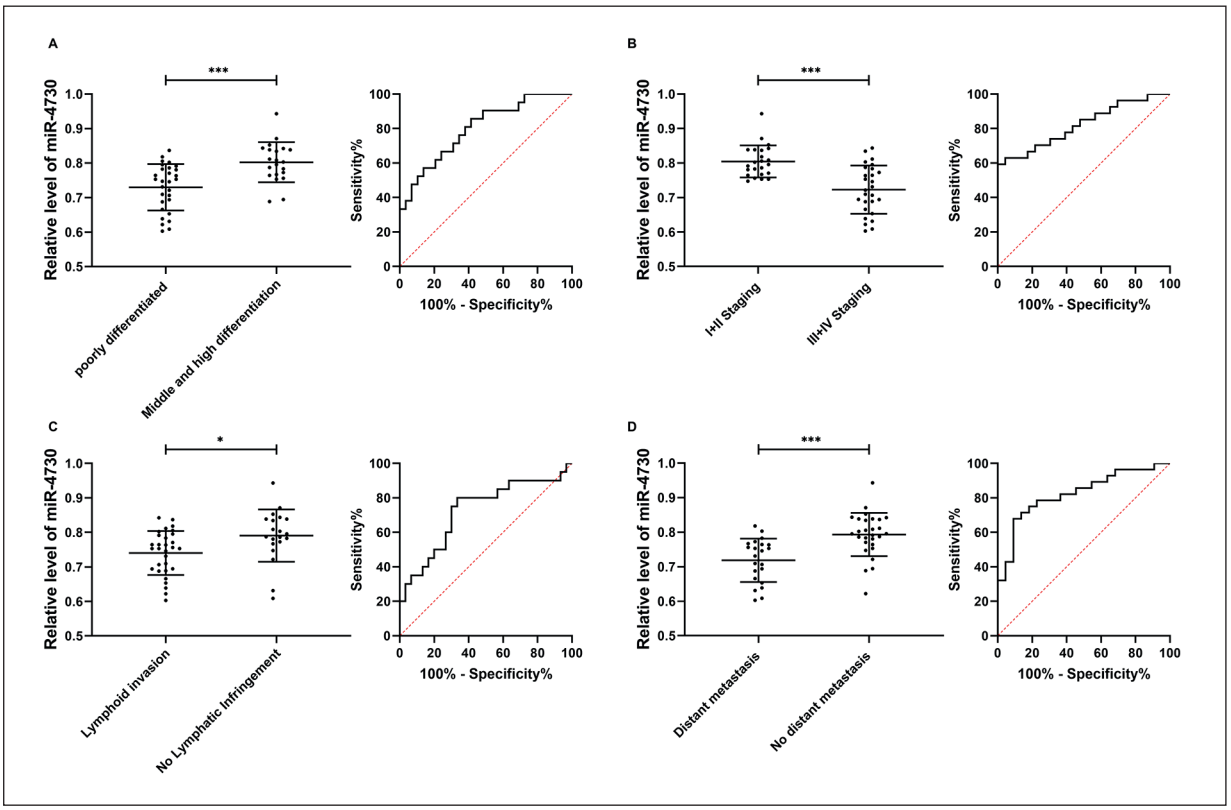

Figure 4. Expression and diagnostic value of miR-4730 for differentiation, TNM staging, lymphatic invasion, and distant metastasis. **A**, The expression and diagnostic value of miR-4730 in patients with different differentiation; **B**, The expression and diagnostic value of miR-4730 in patients with different pathological stages; **C**, The expression and diagnostic value of miR-4730 in patients with and without lymphatic invasion; **D**, The expression and diagnostic value of miR-4730 in patients with and without distant metastases.

Akt/NF-κB signaling pathway transduction, so as to promote the progression and chemotherapy resistance of pancreatic cancer. According to Liang et al[20], miR-33a mediates the decrease of Pim-3 kinase expression and increases the anti-sensitivity of pancreatic cancer cells to gemcitabine. Therefore, in this study, the GEO chip was used to screen out differentially expressed miRs in pancreatic cancer, so as to find out potential diagnostic markers for the disease.

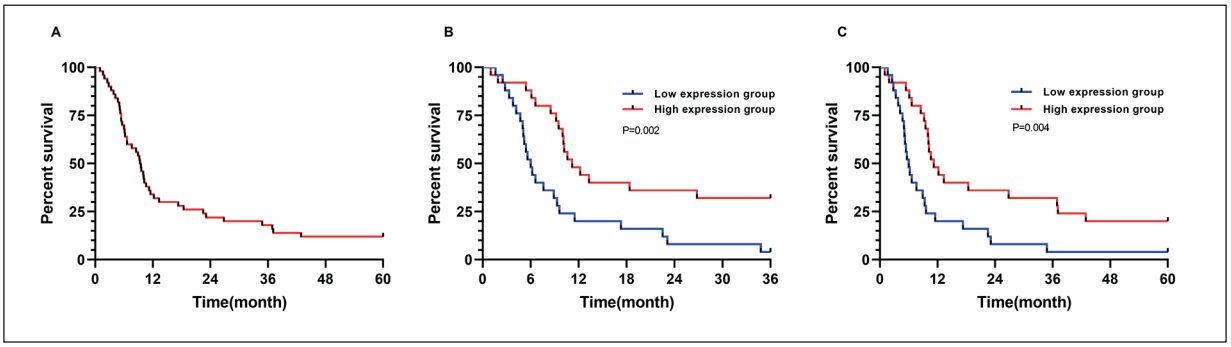

Figure 5. Relationship between miR-4730 and 3- and 5-year survival. **A**, The overall survival rate in Group A; **B**, The 3-year survival rate in the miR-4730 high expression group was higher than that in the miR-4730 low expression group (p=0.002); **C**, The 5-year survival rate in the miR-4730 high expression group was higher than that in the miR-4730 low expression group (p=0.004).

Table V. Univariate and multivariate Cox regression analysis of 3- and 5-year survival.

	Univariate Cox for 3-year survival		Multivariate Cox for 3-year survival		Univariate Cox for 5-year survival		Multivariate Cox for 5-year survival	
Factors	***p*-value**	**HR (95CI%)**	***p*-value**	**HR (95CI%)**	***p*-value**	**HR (95CI%)**	***p*-value**	**HR (95CI%)**
Gender	0.780	1.277 (0.623-2.614)			0.691	0.88 (0.470-1.648)		
Age	0.504	0.939 (0.446-1.975)			0.574	1.186 (0.655-2.146)		
Lesion site	0.868	0.373 (0.143-0.976)			0.793	0.922 (0.501-1.696)		
Differentiation	0.044	4.822 (2.117-10.981)	0.675	0.804 (0.289-2.234)	0.004	0.327 (0.153-0.697)	0.614	0.807 (0.351-1.856)
TNM staging	0.000	0.090 (0.027-0.300)	0.000	13.443 (4.163-43.406)	0.000	3.395 (1.814-6.351)	0.000	8.065 (3.353-19.394)
Lymphatic invasion	0.000	1.175 (0.573-2.408)	0.004	0.121 (0.029-0.500)	0.000	0.178 (0.087-0.363)	0.002	0.240 (0.098-0.590)
Vascular invasion	0.661	0.141 (0.064-0.311)			0.258	1.411 (0.777-2.560)		
Distant metastasis	0.000	0.354 (0.167-0.752)	0.000	0.077 (0.024-0.247)	0.000	0.178 (0.091-0.348)	0.000	0.098 (0.036-0.271)
miR-4730	0.007	0.354 (0.167-0.752)	0.013	3.991 (1.342-11.867)	0.005	0.415 (0.225-0.765)	0.013	3.192 (1.283-7.945)

In this study, differentially expressed miRs in the serum of patients with pancreatic cancer and non-cancer patients were screened out from the GSE112264 chip. The results showed that there were 305 differentially expressed miRs, and that miR-4730 was differentially expressed most, so it was chosen for the detection. miR-4730 belongs to the miR family and is located on human chromosome 17q25.3. Having been rarely studied, miR-4730 is a newly discovered miR in recent years and is only mentioned in screening miR expression profiles of various cancers[21,22]. In this study, the expression of miR-4730 in the cancer tissue and serum of patients with pancreatic cancer was detected. The results showed that the expression in the cancer tissue was significantly lower than that in the adjacent tissue; the expression in the serum was significantly lower in Group A than that in Groups B and C. Huang et al[23] has found that CA199 is differentially expressed in patients with benign pancreatic lesions . In our study, the expression of miR-4730 in the cancer tissue was lower than that in the serum of patients with benign pancreatic lesions. According to the ROC curves, miR-4730 had a good diagnostic value for pancreatic cancer, benign pancreatic lesions, and healthy people, especially for the first and the third ones. The area under the ROC curve of miR-4730 was 0.976, which shows that miR-4730 has a diagnostic value for distinguishing patients with pancreatic cancer from those with benign pancreatic lesions. However, it remains unclear whether there is a correlation between miR-4730 expression and the pathological data of patients with pancreatic cancer. Therefore, further analysis was carried out. The results showed that patients with low miR-4730 expression had poorly differentiated pancreatic cancer, and patients with stages III+IV of pancreatic cancer had higher incidences of lymphatic invasion and distal metastasis. According to the ROC curves, miR-4730 had a high diagnostic value for differentiation, TNM staging, lymphatic invasion, and distal metastasis.

Accordingly, we have preliminarily proved the relationship between miR-4730 expression in pancreatic cancer and the pathological data of patients with the disease. Pancreatic cancer has the lowest 5-year survival rate, and 90% of patients die within 1 year after the disease is detected[24]. Whether there is a correlation of miR-4730 with the patients' survival and prognoses? Can miR-4730 be used as a potential observational index for the survival and prognoses? To answer these questions, we followed up the patients for 5 years, and observed their 3- and 5-year survival. The comparison between the high and low miR-4730 expression groups showed that the 3- and 5-year survival rates in the low expression group were significantly lower than those in the high expression group. This suggests that miR-4730 can be used as a potential observational index for the patients' survival. Finally, the prognostic factors for the survival were analyzed. The results showed that TNM staging, lymphatic invasion, distal metastasis, and miR-4730 were the independent prognostic factors. Previous researches have shown that TNM staging, lymphatic invasion, and distal metastasis are independent prognostic factors affecting the patients' prognoses[25,26], but our study is the first report that miR-4730 can be used as the independent prognostic factor. The clinical value of miR-4730 in pancreatic cancer is preliminarily determined in this study, so it is expected to become a potential diagnostic and prognostic indicator for the disease.

However, there are still some limitations. First, the relevant mechanism of miR-4730 in pancreatic cancer is unclear in this clinical study. Second, the expression of serum miR-4730 after treatment was not detected, so whether miR-4730 is differentially expressed in the patients' serum after treatment needs further investigation. Therefore, we hope to detect the expression of serum miR after treatment in subsequent studies. Additionally, basic experiments should be carried out to explore the pathway through which miR-4730 affects the development and progression of pancreatic cancer, so as to supplement our research results.

Conclusions

In summary, for patients with pancreatic cancer, those with low miR-4730 expression have poor survival and prognoses, so miR-4730 can be used as a potential observational index for the prognosis and diagnosis of the disease.

Conflict of Interest

The Authors declare that they have no conflict of interests.

Supportive Foundations

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

References

1) Kamisawa T, Wood LD, Itoi T, Takaori K. Pancreatic cancer. Lancet 2016; 388: 73-85.
2) Kleeff J, Korc M, Apte M, La Vecchia C, Johnson CD, Biankin AV, Neale RE, Tempero M, Tuveson DA, Hruban RH, Neoptolemos JP. Pancreatic cancer. Nat Rev Dis Primers 2016; 2: 16022.
3) Ilic M, Ilic I. Epidemiology of pancreatic cancer. World J Gastroenterol 2016; 22: 9694-9705.
4) Suker M, Beumer BR, Sadot E, Marthey L, Faris JE, Mellon EA, El-Rayes BF, Wang-Gillam A, Lacy J, Hosein PJ, Moorcraft SY, Conroy T, Hohla F, Allen P, Taieb J, Hong TS, Shridhar R, Chau I, van Eijck CH, Koerkamp BG. FOLFIRINOX for locally advanced pancreatic cancer: a systematic review and patient-level meta-analysis. Lancet Oncol 2016; 17: 801-810.
5) Yam BL, Siegelman ESJMMOGC. The role of diagnostic radiology in pancreatic cancer management. 2016: 189.
6) Hua Y, Chen H, Wang L, Wang F, Wang P, Ning Z, Li Y, Liu L, Chen Z, Meng Z. Low serum miR-373 predicts poor prognosis in patients with pancreatic cancer. Cancer Biomark 2017; 20: 95-100.
7) O'Brien DP, Sandanayake NS, Jenkinson C, Gentry-Maharaj A, Apostolidou S, Fourkala EO, Camuzeaux S, Blyuss O, Gunu R, Dawnay A, Zaikin A, Smith RC, Jacobs IJ, Menon U, Costello E, Pereira SP, Timms JF. Serum CA19-9 is significantly upregulated up to 2 years before diagnosis with pancreatic cancer: implications for early disease detection. Clin Cancer Res 2015; 21: 622-631.
8) Lin S, Gregory RI. MicroRNA biogenesis pathways in cancer. Nat Rev Cancer 2015; 15: 321-333.
9) Hoffman Y, Bublik DR, Ugalde AP, Elkon R, Biniashvili T, Agami R, Oren M, Pilpel Y. 3'UTR shortening potentiates microRNA-based repression of pro-differentiation genes in proliferating human cells. PLoS Genet 2016; 12: e1005879.
10) Zhang L, Yang CS, Varelas X, Monti S. Altered RNA editing in 3' UTR perturbs microRNA-mediated regulation of oncogenes and tumor-suppressors. Sci Rep 2016; 6: 23226.
11) Rupaimoole R, Slack FJ. MicroRNA therapeutics: towards a new era for the management of cancer and other diseases. Nat Rev Drug Discov 2017; 16: 203-222.
12) Cheng CJ, Bahal R, Babar IA, Pincus Z, Barrera F, Liu C, Svoronos A, Braddock DT, Glazer PM, Engelman DM, Saltzman WM, Slack FJ. MicroRNA silencing for cancer therapy targeted to the tumour microenvironment. Nature 2015; 518: 107-110.
13) Schlitter AM, Jesinghaus M, Jager C, Konukiewitz B, Muckenhuber A, Demir IE, Bahra M, Denkert C, Friess H, Kloeppel G, Ceyhan GO, Weichert W. pT but not pN stage of the 8th TNM classification significantly improves prognostication in pancreatic ductal adenocarcinoma. Eur J Cancer 2017; 84: 121-129.
14) Neoptolemos JP, Kleeff J, Michl P, Costello E, Greenhalf W, Palmer DH. Therapeutic developments in pancreatic cancer: current and future perspectives. Nat Rev Gastroenterol Hepatol 2018; 15: 333-348.
15) Allemani C, Matsuda T, Di Carlo V, Harewood R, Matz M, NikšiĐ M, Bonaventure A, Valkov M, Johnson CJ, Estève J. Global surveillance of trends in cancer survival: analysis of individual records for 37,513,025 patients diagnosed with one of 18 cancers during 2000–2014 from 322 population-based registries in 71 countries (CONCORD-3). 2017.
16) Yamamoto T, Yagi S, Kinoshita H, Sakamoto Y, Okada K, Uryuhara K, Morimoto T, Kaihara S, Hosotani R. Long-term survival after resection of pancreatic cancer: a single-center retrospective analysis. World J Gastroenterol 2015; 21: 262-268.
17) Mao X, Wang L. Clinical application value of serum miR-21 and CA199 in the diagnosis of pancreatic cancer. Chongqing Medicine 2018; 47: 2319-2322.
18) Abue M, Yokoyama M, Shibuya R, Tamai K, Yamaguchi K, Sato I, Tanaka N, Hamada S, Shimosegawa T, Sugamura K, Satoh K. Circulating miR-483-3p and miR-21 is highly expressed in plasma of pancreatic cancer. Int J Oncol 2015; 46: 539-547.
19) Li J, Wu H, Li W, Yin L, Guo S, Xu X, Ouyang Y, Zhao Z, Liu S, Tian Y, Tian Z, Ju J, Ni B, Wang H. Downregulated miR-506 expression facilitates pancreatic cancer progression and chemoresistance via SPHK1/Akt/NF-kappaB signaling. Oncogene 2016; 35: 5501-5514.
20) Liang C, Yu XJ, Guo XZ, Sun MH, Wang Z, Song Y, Ni QX, Li HY, Mukaida N, Li YY. MicroRNA-33a-mediated downregulation of Pim-3 kinase expression renders human pancreatic cancer cells sensitivity to gemcitabine. Oncotarget 2015; 6: 14440-14455.
21) Yata K, Beder LB, Tamagawa S, Hotomi M, Hirohashi Y, Grenman R, Yamanaka N. MicroRNA expression profiles of cancer stem cells in head and neck squamous cell carcinoma. Int J Oncol 2015; 47: 1249-1256.
22) Yokoi A, Matsuzaki J, Yamamoto Y, Yoneoka Y, Takahashi K, Shimizu H, Uehara T, Ishikawa M, Ikeda SI, Sonoda T, Kawauchi J, Takizawa S, Aoki Y, Niida S, Sakamoto H, Kato K, Kato T, Ochiya T. Integrated extracellular microRNA profiling for ovarian cancer screening. Nat Commun 2018; 9: 4319.
23) Huang S, Chong H, Zhang Y, Lan XJJoNM. The value of 18F-FDG PET/CT in the differential diagnosis of pancreatic lesions: comparison with CA19-9, enhanced CT or enhanced MR. 2018; 59: 153-153.
24) Siegel RL, Miller KD, Jemal A. Cancer statistics, 2015. CA Cancer J Clin 2015; 65: 5-29.
25) Nitta T, Nakamura T, Mitsuhashi T, Asano T, Okamura K, Tsuchikawa T, Tamoto E, Murakami S, Noji T, Kurashima Y, Ebihara Y, Nakanishi Y, Shichinohe T, Hira-

NO S. The impact of margin status determined by the one-millimeter rule on tumor recurrence and survival following pancreaticoduodenectomy for pancreatic ductal adenocarcinoma. Surg Today 2017; 47: 490-497.

26) DENG T, YUAN Y, ZHANG C, ZHANG C, YAO W, WANG C, LIU R, BA Y. Identification of circulating miR-25 as a potential biomarker for pancreatic cancer diagnosis. Cell Physiol Biochem 2016; 39: 1716-1722.