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Expression and clinical diagnostic value of miR-383 in patients with severe preeclampsia

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Abstract: This study aimed to investigate the expression and clinical diagnostic value of miR-383 in patients with severe preeclampsia. Thirty patients with severe preeclampsia from July 2017 to December 2018 were selected as a research group, twenty healthy pregnant women undergoing physical examination at the same period were selected as a control group, and miR-383 and miR-16 in placenta tissue of the two groups were detected by qRT-PCR. ROC curve was drawn to evaluate the predictive value of diagnostic efficiency, Spearman test was used for correlation analysis, and Logistic univariate and multivariate analysis was performed on the risk factors related to the metastasis of severe preeclampsia. The miR-383 expression in the research group was significantly lower than that in the control group (P< 0.001), while the miR-16 expression in the research group was significantly higher than that in the control group (P< 0.001). The miR-383 and miR-16 expression levels were tied to TNM staging and metastasis (P< 0.001). The sensitivity, specificity and AUC of miR-383 single diagnosis were 75.00%, 83.33% and 0.847 respectively, and those of miR-16 single diagnosis were 65.00%, 63.33% and 0.728 respectively. The relative expression of miR-383 in placenta tissue was negatively correlated with APACHE II score of severe preeclampsia (r = -0.4129, P= 0.0233), but the relative expression of miR-16 in placenta tissue was positively correlated with APACHE II score of severe preeclampsia (r = 0.9833, P< 0.001). Blood pressure, miR-383, miR-16 at the admission of pregnant women were independent risk factors for severe preeclampsia. miR-383 and miR-16 might participate in the process of occurrence, development and metastasis of severe preeclampsia, and could be used as potential biomarkers of placental tissue for its diagnosis and disease assessment of metastasis.

Key words: miR-383; Severe preeclampsia; Clinical diagnosis; miR-16; APACHE II score.

Introduction

Preeclampsia is a kind of pregnancy disease with great harm. With the increase of gestational weeks of patients, pregnant women and fetuses bear more and more harm (1, 2). Although the morbidity of severe preeclampsia is relatively low, it has increased by 2% year by year (3). According to the disease severity of pregnant women, preeclampsia can be divided into mild and severe ones. Inadequate vascular remodeling of the placenta bed and systemic vascular endothelial injury are all the causes leading to adverse pregnancy outcomes in preeclampsia patients. Severe preeclampsia is often accompanied by impairment of the functions of important organs of the heart, brain and kidney, which seriously threatens the health of mothers and infants. At the same time, it places great health and economic burden on the family of pregnant women. In order to reduce the harm of severe preeclampsia, attention should be paid to the monitoring of early pregnancy or early preeclampsia (4, 5). Measurement of early blood pressure and albuminuria detection are the conventional detection indicators for preeclampsia patients in clinical practice (6, 7), but the initial cause of severe preeclampsia is still unclear. With the continuous development of molecular biology, medical researchers began to analyze the pathogenesis of human severe preeclampsia from the direction of gene biology of the disease. However, inhibiting the progress of severe preeclampsia is an important part of the current study (8).

The current research showed that MicroRNAs (miRNAs) were an endogenous non-coding single-stranded RNA with gene expression regulation function (9, 10), and it inhibits or promotes diseases in different diseases, and it abnormally expresses in severe preeclampsia. However, miRNAs with different expression levels detected in different studies are different (11).

At present, there is a lack of research on the expression and predictive value of miR-383 and miR-16 in severe preeclampsia metastasis. However, it has been reported that the miR-16 expression is up-regulated in severe preeclampsia cells, and its appropriate down-regulation can inhibit the progression of severe preeclampsia (12). On the contrary, miR-383 has been proved to be far less expressed in colorectal cancer, medulloblastoma and other tumor tissues than in normal tissues. Up-regulating the miR-383 expression can inhibit the proliferation of colon cancer cells and reduce the invasion and migration (13). We speculated that miR-383 and miR-16 were related to the progression of preeclampsia. Therefore, in order to provide a new theoretical basis for the diagnosis and treatment of se-

vere preeclampsia in molecular biology, the expression characteristics and clinical significance of miR-383 and miR-16 were studied.

Materials and Methods

Data collection

Thirty patients with severe preeclampsia treated in The Second Affiliated Hospital of Nanhua University from July 2017 to December 2018 were selected as the research group, and 20 healthy pregnant women who underwent physical examination during the same period were selected as the control group. They were (25.45±3.03) and (25.27±3.19) years old on average. Inclusion criteria: All the patients were in accordance with the diagnosis guidelines of the expert committee of severe preeclampsia; all of them were primiparas, with 29-41 weeks of gestation; they were conscious (14); they had normal liver and kidney function and no other malignant tumors. Exclusion criteria: All patients who received chemotherapy, immunotherapy and radiotherapy before surgery were excluded. The research was approved by the Ethics Committee of the Second Affiliated Hospital of Nanhua University, patients and their families were informed in advance, and they signed informed consent forms.

Main reagents, instruments and detection methods

Main reagents and instruments

Main reagents and instruments were as follows: Trizol reagent (Applied Invitrogen, U.S.), qRT-PCR kit and minScript reverse transcription kit (Dalian TaKaRa company), HBS-1096A enzyme label analyzer (Nanjing Detie Experimental Equipment Co., Ltd.), real-time quantitative PCR instrument (BioRad company, U.S.), and primer sequences of miR-383 and miR-16 as well as internal reference U6 and miRNA negative control were synthesized and designed by Shanghai GenePharma Company (Table 1).

Detection of miR-383 and miR-16

The expression levels of miR-383 and miR-16 of placenta tissue in the two groups were detected by qRT-PCR. Total RNA of placental tissue was extracted according to Trizol reagent operation instructions and dissolved in 20 μL DEPC water, and it was then reverse-transcribed using a reverse transcription kit. The reaction system was as follows: M-MLV 1 μl, Oligo (dT) 1 μl, RNA enzyme inhibitor 0.5 μl, d NTPs 1 μl, RNAse free water 15 μl. After that, it was incubated 60 min at 38 ℃. We took 1 μl c DNA and put it at 85 ℃ for 5 s, and we used the synthesized c DNA as a template for qRT-PCR amplification. The PCR reaction system was prepared: 10×PCR buffer 2.5 μl, d NTPs 1 μl, upstream and downstream primers 1 μl each, Taq DNA Polymerase 0.25 μl, ddH2O supplemented to 25 μl. Reaction conditions were as follows: pre-denaturation 95 ℃, 15 min, denaturation 95 ℃, 15 s, annealing 60 ℃, 30 s, a total of 35 cycles, an extension for 15 min at 72 ℃. Each sample was provided with 3 multiple wells for 3 repeated tests, and U6 was regarded as the internal reference in miR-383 and miR-16. Afterward, the amplification curve and melting curve of Real-Time PCR were confirmed, and the relative amount of the target gene was calculated based on the result parameters. The relative quantification of the target gene was calculated by $2^{-\triangle CT}$.

Statistical methods

SPSS 17.0 software system was used for statistical analysis, the counting data were expressed by the number of cases/percentages [n(%)], and χ^2 test was used for comparison between the two groups. The measurement data were expressed by mean number (X±sd), and the comparison between groups was conducted by a t-test or F test. ROC curve was drawn to select the maximum point cut-off value of the Youden index and to evaluate the predictive value of diagnostic efficiency in miR-383 and miR-16 expression in placenta tissue. Spearman test was used for correlation analysis, and Logistic univariate and multivariate analysis was performed on the risk factors related to the metastasis of moderate and severe preeclampsia in patients with severe preeclampsia. A p-value lower than 0.05 was considered statistically significant.

Results

General clinical data of patients

The age, number of pregnancies, APACHE II score, lesion site and lymph node metastasis of the research group and the control group were compared, and there was no significant difference between the two groups in terms of age, gender and other clinical general data (P> 0.05) based on Table 2.

Expression levels of miR-383 and miR-16 in research group and control group

The results of qRT-PCR showed that the miR-383 expression in placenta tissue of the research group and the control group were (0.40±0.15) and (0.60±0.12) respectively, the miR-16 expression in placenta tissue of the two groups were (3.15±1.13) and (1.38±1.13) respectively. Compared with both groups, the miR-383 expression in the research group was significantly lower than that in the placenta tissue of the control group, with a statistically significant difference (P< 0.001). However, the miR-16 expression in the research group was significantly higher than that in the control group (P< 0.001, Figures 1A & 1B).

Relationship between the expression levels of miR-383 and miR-16 and clinicopathological characteristics of severe preeclampsia

The expression levels of miR-383 and miR-16 had

Table 1. Primer sequences of miR-383, miR-16 and internal reference U6.

Group	Upstream primer	Downstream primer
miR-383	5'-GTGCAGGGTCCGAGGT-3'	5'-AGATCAGAAGGTGATTGTGGCT-3'
miR-16	5'-TAGCAGCACGTAAATATTGGCG-3'	5'-TGCGTGTCGTG GAGTC-3'
U6	5'-CTCGCTTCGGCAGCACA-3'	5'-AACGCTTCACGAATTTGCGT-3'

Table 2. General clinical data of patients in current research.

Factor	Research group (n=30)	Control group (n=20)	t/X²	P
Age (years)			0.061	0.805
≤25	10 (33.33)	6 (30.00)		
>25	20 (66.67)	14 (70.00)		
Number of pregnancies			0.066	0.797
Multipara	8 (26.67)	6 (30.00)		
Primipara	22 (73.33)	14 (70.00)		
Gestational weeks of onset (week)			-	-
≤27	14 (46.67)	-		
>27	16 (53.33)	-		
Hemoglobin of pregnant women at admission			0.000	1.000
Normal	6 (20.00)	4 (20.00)		
Abnormal	24 (80.00)	16 (80.00)		
Blood pressure of pregnant women at admission			0.178	0.673
Normal	8 (26.67)	2 (20.00)		
Hypertension	22 (73.33)	8 (80.00)		
Polyembryony			0.042	0.838
Yes	8 (26.67)	3 (30.00)		
No	22 (73.33)	7 (70.00)		
Blood pressure of pregnant women at the end of pregnancy			0.833	0.361
Normal	23 (76.67)	9 (90.00)		
Hypertension	7 (33.33)	1 (10.00)		
APACHE II score			-	-
<9	11 (36.67)	-		
9-14	9 (30.00)	-		
>14	10 (33.33)	-		

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Factor	Research group (n=30)	Control group (n=20)	t/X²	P
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≤25	10 (33.33)	6 (30.00)		
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Primipara	22 (73.33)	14 (70.00)		
Gestational weeks of onset (week)			-	-
≤27	14 (46.67)	-		
>27	16 (53.33)	-		
Hemoglobin of pregnant women at admission			0.000	1.000
Normal	6 (20.00)	4 (20.00)		
Abnormal	24 (80.00)	16 (80.00)		
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Normal	23 (76.67)	9 (90.00)		
Hypertension	7 (33.33)	1 (10.00)		
APACHE II score			-	-
<9	11 (36.67)	-		
9-14	9 (30.00)	-		
>14	10 (33.33)	-		

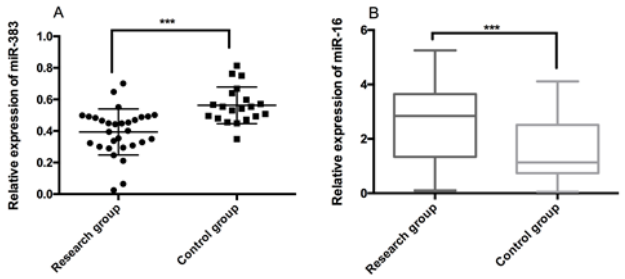

Figure 1. Comparison of relative expression results of miR-383 and miR-16 in placenta tissue of the research group and control group. **A:** The miR-383 expression in the research group was significantly lower than that in the control group (P< 0.001). **B:** The miR-16 expression in the research group was significantly higher than that in the control group (P< 0.001). Note: *** indicated P< 0.001.

nothing to do with the age of patients with severe preeclampsia, the number of pregnancies, the gestational weeks of onset, hemoglobin and polyembryony of pregnant women at admission, but was concerned with APACHE II score, blood pressure of pregnant women at admission (Table 3).

Correlation of miR-383 and miR-16 in severe preeclampsia with different APACHE II scores

The expression level of miR-383 in the placental tissue of patients with APACHE II scores < 9, 9-14 and > 14 of different severe preeclampsia were (0.50±0.13), (0.39±0.15) and (0.32±0.15), respectively. The expression level of miR-16 in the placental tissue of patients with APACHE II scores < 9, 9-14 and > 14 of different severe preeclampsia were (2.03±1.15), (3.03±1.15) and (4.25±1.15), respectively. Compared with patients with APACHE II score < 9, the relative expression levels of miR-383 and miR-16 in the placenta tissue of patients 9-14 and > 14 decreased significantly (P < 0.05). With the increase of the APACHE II score, the relative expression of miR-383 in placenta tissue decreased, while the relative expression of miR-16 in placenta tissue increased continuously. The APACHE II score of patients < 9 was set to 1, 9-14 was set to 2, and > 14 was set to 3. Spearman correlation analysis between the relative expression levels of miR-383 and miR-16 in placenta tissue and APACHE II score of different severe preeclampsia indicated that the relative expression of miR-383 in placenta tissue was negatively correlated with APACHE II score of severe preeclampsia (r = -0.4129, P = 0.0233), and the relative expression of miR-16 in placenta tissue was positively correlated with APACHE II score of severe preeclampsia (r = 0.9833, P< 0.001) based on Figures 2 A-D.

Diagnostic value of miR-383 and miR-16 in the prognosis of severe preeclampsia

In diagnosing severe preeclampsia, the sensitivity, specificity and AUC of miR-383 single diagnosis were 75.00, 83.33, and 0.847 respectively, and the sensitivity, specificity and AUC values of miR-16 single diagnosis were 65.00, 63.33, and 0.728 respectively. (Table 4, Figures 3A and 3B).

Prognostic value of miR-383 and miR-16 in patients with severe preeclampsia

Prognosis of patients with severe preeclampsia after treatment

According to the complications of patients with severe preeclampsia after treatment, they were divided into a good prognosis group and poor prognosis group.

Table 3. Relationship between the expression levels of miR-383 and miR-16 clinicopathological characteristics of severe preeclampsia.

Group	n	miR-383	t/F	P	miR-16	t/F	P
Age (years)			0.000	0.999		0.0696	0.945
≤ 25	10	0.40±0.10			3.10±1.14		
> 25	20	0.40±0.20			3.13±1.10		
Number of pregnancies			0.145	0.886		0.021	0.983
Multipara	8	0.39±0.16			3.14±1.14		
Primipara	22	0.38±0.17			3.15±1.13		
Gestational weeks of onset (week)			1.957	0.060		0.618	0.542
≤ 27	14	0.45±0.15			2.90±1.16		
> 27	16	0.35±0.13			3.16±1.14		
Hemoglobin of pregnant women at admission			1.899	0.068		1.702	0.100
Normal	6	0.46±0.15			2.70±1.15		
Abnormal	24	0.33±0.15			3.60±1.16		
Blood pressure of pregnant women at admission			3.398	0.002		2.207	0.036
Normal	8	0.50±0.15			2.80±1.16		
Hypertension	22	0.30±0.14			3.85±1.15		
Polyembryony			0.159	0.875		0.021	0.983
Yes	8	0.40±0.16			3.14±1.15		
No	22	0.39±0.15			3.15±1.13		
Blood pressure of pregnant women at the end of pregnancy			2.607	0.015		2.182	0.038
Normal	23	0.48±0.14			2.60±1.10		
Hypertension	7	0.32±0.15			3.64±1.12		
APACHE II score			4.245	0.025		9.768	0.001
< 9	11	0.50±0.13			2.03±1.15		
9-14	9	0.39±0.15			3.03±1.15		
> 14	10	0.32±0.15			4.25±1.15		

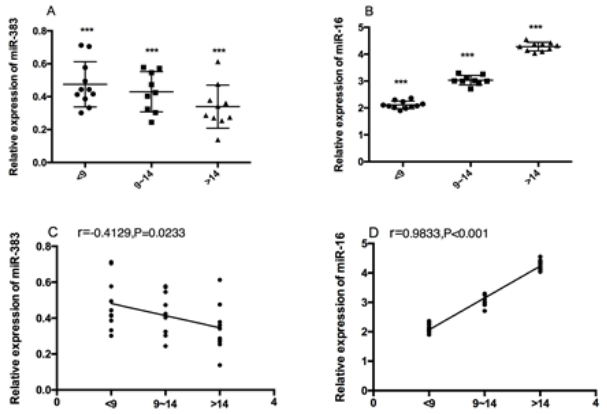

Figure 2. Correlation between the relative expression levels of miR-383 and miR-16 in placenta tissue of the research group and APACHE II score of severe preeclampsia. **A.** With the increase of the APACHE II score in severe preeclampsia, the relative expression of miR-383 in placenta tissue decreased (P< 0.05). Note: * indicated that 9-14 was compared with > 14 (P< 0.05), and *** indicated that compared with < 9, stages IV and II, and > 14, P was less than 0.001. **B.** With the increase of the APACHE II score in severe preeclampsia, the relative expression of miR-16 in placenta tissue decreased (P< 0.05). Note: * indicated that 9-14 was compared with > 14 (P< 0.05), and *** indicated that compared with < 9, stages IV and II, and > 14, P was less than 0.001. **C.** The relative expression of miR-383 in placenta tissue was negatively correlated with the APACHE II score of severe preeclampsia (r = -0.4129, P = 0.0233). **D.** The relative expression of miR-16 in placenta tissue was positively correlated with APACHE II score of severe preeclampsia (r = 0.9833, P< 0.001).

Compared with the two groups, the probability of postpartum complications in the good prognosis group was significantly lower than that in the poor prognosis group (P< 0.05, Table 5).

Univariate analysis of prognosis and related factors of severe preeclampsia

Logistic univariate analysis of risk factors related to the metastasis of moderate and severe preeclampsia in patients with severe preeclampsia showed that there were significant differences in blood, APACHE II score, miR-383, miR-16 between pregnant women with metastasis of severe preeclampsia and those without metastasis of severe preeclampsia at admission (P< 0.05). The age, lesion site, APACHE II score, miR-383, miR-16 of patients were all related to metastasis of severe preeclampsia and were risk factors for metastasis of severe preeclampsia (Tables 6 and 7).

Multivariate analysis of metastasis of severe preeclampsia and related factors

Risk factors related to the prognosis of severe preeclampsia were analyzed by multivariate conditional Logistic regression, and the results revealed that blood pressure, miR-383 and miR-16 at admission were independent risk factors for metastasis of severe preeclampsia (Table 8).

Discussion

miRNA is ubiquitous in eukaryotes and plays a role in gene regulation in cell biological processes such as cell development, differentiation and apoptosis, and the biological characteristics of different miRNA are different (15). The differential expression of miRNA in the placental tissue of preeclampsia and normal pregnant women was found in some functional studies of miRNA (16). Severe preeclampsia is an important factor leading to poor treatment effects and pregnancy outcomes of

Table 4. Relationship between the expression of miR-16 and clinicopathological characteristics of severe preeclampsia.

Group	n	miR-16	t/F	P
Age (years)			0.0696	0.945
≤ 25	10	3.10±1.14		
> 25	20	3.13±1.10		
Number of pregnancies			0.021	0.983
Multipara	8	3.14±1.14		
Primipara	22	3.15±1.13		
Gestational weeks of onset (week)			0.618	0.542
≤ 27	14	2.90±1.16		
> 27	16	3.16±1.14		
Hemoglobin of pregnant women at admission			1.702	0.100
Normal	6	2.70±1.15		
Abnormal	24	3.60±1.16		
Blood pressure of pregnant women at admission			2.207	0.036
Normal	8	2.80±1.16		
Hypertension	22	3.85±1.15		
Polyembryony			0.021	0.983
Yes	8	3.14±1.15		
No	22	3.15±1.13		
Blood pressure of pregnant women at the end of pregnancy			2.182	0.038
Normal	23	2.60±1.10		
Hypertension	7	3.64±1.12		
APACHE II score			9.768	0.001
< 9	11	2.03±1.15		
9-14	9	3.03±1.15		
> 14	10	4.25±1.15		

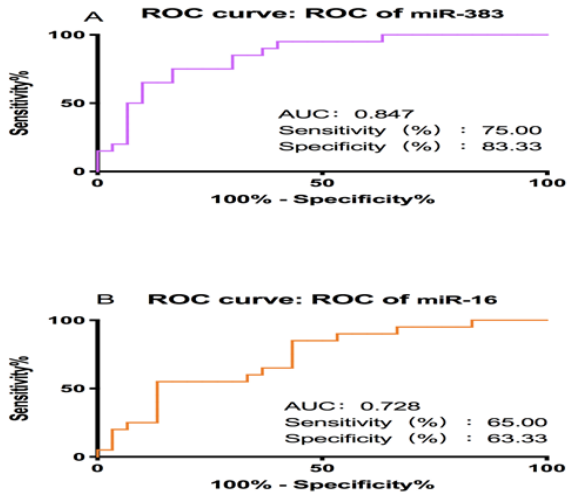

Figure 3. ROC curve of miR-383 and miR-16 in placental tissue for the diagnosis of severe preeclampsia. **A:** The sensitivity and specificity of miR-383 in placenta tissue in diagnosing severe preeclampsia were 75.00 and 83.33%, respectively. **B:** The sensitivity and specificity of miR-16 in placenta tissue in diagnosing severe preeclampsia were 65.00 and 63.33%, respectively.

preeclampsia patients. Diagnosis and treatment of patients with severe preeclampsia could reduce the risks of maternal death during pregnancy, premature delivery and neonatal death. Therefore, finding miRNA biomarkers closely related to the diagnosis and treatment of severe preeclampsia has important clinical significance (17).

In this study, we first detected the expression differences of miR-383 and miR-16 in the placenta tissue of patients with severe preeclampsia and healthy people by qRT-PCR. The results showed that the miR-383 expression in the research group was significantly lower than that in the control group and there was a statistically significant difference, while the miR-16 expression in the research group was significantly higher than that in the control group and there was a statistically significant difference. miRNA could affect the biological function of tumor cells by regulating gene expression and regulating related tumor signal pathways, thus affecting the development of tumors (18). miR-383 down-regulated in colorectal cancer, hepatocellular carcinoma and other diseases, and up-regulation of miR-383 could promote apoptosis of colorectal cancer cells (19, 20).

Researchers verified that miR-383 down-regulated the VEGFA expression and inhibited the angiogenesis of tumor endothelial cells by targeting the 3'-UTR of the VEGFA gene to inhibit its translation efficiency (21, 22). However, miR-16, as a potential target molecule for treating severe preeclampsia, was abnormally elevated in a variety of tumor tissues including severe preeclampsia. Moreover, some researches believed that miR-16 could promote the growth of placental decidua derived mesenchymal stem cells of preeclampsia patients by inhibiting miR-16 expression (23, 24). Hence, we believed that miR-383 expression down-regulated in the placenta tissue of patients with severe preeclampsia. On the contrary, miR-16 up-regulated in the placenta tissue of patients with severe preeclampsia. Then we started with the clinical data of patients in the research group to analyze the relationship between the expression levels of miR-383 and miR-16 and the clinicopathological characteristics of severe preeclampsia. Based

Table 5. Diagnostic value of miR-383 and miR-16 in treatment of placenta tissue for the efficacy of PD patients.

Indicators	miR-383	miR-16
AUC	0.847	0.728
95%CI	0.7388-0.9545	0.5869-0.8697
Std. Error	0.0550	0.0721
Cut-off value	0.493	2.212
Sensitivity (%)	75.00	65.00
Specificity (%)	83.33	63.33

Table 6. Incidence rate of complications.

Group	The good prognosis (n=20)	Poor prognosis (n=10)	X^2	P
Pulmonary edema	0 (0.00)	1 (10.00)	-	-
Acute renal insufficiency	1 (5.00)	2 (20.00)	-	-
HELLP syndrome	1 (5.00)	1 (10.00)	-	-
Cardiovascular and cerebrovascular accident	0 (0.00)	1 (10.00)	-	-
Total complication rate	2 (10.00)	5 (50.00)	5.963	0.015

Table 7. Assignment description of factors related to metastasis of severe preeclampsia.

Correlative factor	Assignment description
Age (years)	< 25 =0; ≥ 25 =1
Number of pregnancies	Primipara = 0; multipara =1
Gestational weeks of onset (week)	≤ 27=0; > 27=1
Hemoglobin of pregnant women at admission	Normal = 0; abnormal =1
Blood pressure of pregnant women at admission	Normal = 0; hypertension =1
Polyembryony	Yes = 0; no =1
Blood pressure of pregnant women at the end of pregnancy	Normal = 0; hypertension =1
miR-383	< 0.451=0; > 0.451=1
miR-16	< 4.841=0; > 4.841=1
APACHE II score	< 9 + 9-14=0; > 14=1

Table 8. Univariate analysis of metastasis of severe preeclampsia and related factors.

Factor	The good prognosis (n=20)	Poor prognosis (n=10)	X^2	P
Age (years)			0.075	0.784
≤25	7 (35.00)	3 (30.00)		
>25	13 (65.00)	7 (70.00)		
Number of pregnancies			0.085	0.770
Multipara	5 (25.00)	3 (30.00)		
Primipara	15 (75.00)	7 (70.00)		
Gestational weeks of onset (week)			0.268	0.605
≤27	10 (50.00)	4 (40.00)		
>27	10 (50.00)	6 (60.00)		
Hemoglobin of pregnant women at admission			0.000	1.000
Normal	4 (20.00)	2 (20.00)		
Abnormal	16 (80.00)	8 (80.00)		
Blood pressure of pregnant women at admission			5.455	0.020
Normal	8 (40.00)	0 (0.00)		
Hypertension	12 (60.00)	10 (100.00)		
Polyembryony			1.148	0.284
Yes	6 (30.00)	5 (50.00)		
No	14 (70.00)	5 (50.00)		
Blood pressure of pregnant women at the end of pregnancy			18.260	< 0.001
Normal	20 (100.00)	3 (30.00)		
Hypertension	0 (0.00)	7 (70.00)		
miR-383	0.64±0.30	0.30±0.16	4.179	< 0.001
miR-16	6.38±2.35	4.18±1.85	3.295	0.002
APACHE II score			9.459	0.009
<9	10 (50.00)	1 (10.00)		
9-14	7 (35.00)	2 (20.00)		
>14	3 (15.00)	7 (70.00)		

on the analysis results, we speculated that the expression levels of miR-383 and miR-16 were linked to the APACHE II score of severe preeclampsia, and the blood pressure of pregnant women at admission. At present, although there is no specific study on the pathological characteristics of patients with severe preeclampsia as well as miR-383 and miR-16, there are reports on miRNAs and severe preeclampsia, which suggest that the miR-125b-1-3p expression detected by qRT-PCR shows a decreasing trend in placenta tissue of preeclampsia patients. Furthermore, the miR-125b-1-3p expression in patients with different preeclampsia severity was further detected through experiments, and we discovered that the lower its expression in placenta tissue was, the more severe state of illness was, indicating that the expression change of miRNA was tied to the clinicopathological staging of severe preeclampsia, which was similar to the results of this study, and was an excellent supplement (25).

Then, we analyzed the correlation between miR-383 and miR-16 in severe preeclampsia with different APACHE II scores and found that the relative expression of miR-383 in placenta tissue decreased with the increase of APACHE II scores, while the relative expression of miR-16 in placenta tissue increased continuously. Spearman correlation analysis signified that the relative expression of miR-383 in placenta tissue was negatively correlated with APACHE II score in severe preeclampsia, and the relative expression of miR-16 in placenta tissue was positively correlated with APACHE II score in severe preeclampsia. miRNAs were proved to be strongly linked to disease condition assessment. Through inhibiting or promoting disease development in different tumors, the expression level of miRNAs also increased or decreased significantly with disease progression (26). Finally, we analyzed the diagnostic value of miR-383 and miR-16 in metastasis of severe preeclampsia and the predictive value for metastasis of severe preeclampsia. By drawing ROC curves, we discovered that the single diagnosis of miR-383 and miR-16 had better sensitivity, specificity and AUC; in diagnosing severe preeclampsia patients, liver and kidney function, blood and urine routine, coagulation indicator and ultrasonic cardiogram and other related examinations were all routine clinical auxiliary examinations. There was a certain degree of misdiagnosis and missed diagnosis rate for metastatic severe preeclampsia in vivo, and combination with a placental tissue tumor marker could better improve the diagnostic efficiency (27, 28).

Through Logistic univariate and multivariate analysis of risk factors related to the prognosis of severe preeclampsia patients with moderate and severe preeclampsia, we confirmed that APACHE II score, blood pressure of pregnant women at admission, miR-383 and miR-16 were independent risk factors for metastasis of severe preeclampsia. However, the diagnostic efficacy and predictive value of miR-383 and miR-16 expression changes in the placenta tissue of patients with severe preeclampsia metastasis have not been studied before. In this study, miR-383 and miR-16 have certain predictive value for the diagnosis and disease prognosis in metastasis of patients with severe preeclampsia (29-44).

This study confirmed the expression and predictive value of miR-383 and miR-16 in patients with severe preeclampsia, but there are some limitations. For ins-

tance, this study did not specifically analyze the regulatory effect of miR-383 and miR-16 expression changes on severe preeclampsia, and further explain its biological function. Besides, miR-383 and miR-16 were not analyzed with clinical routine tumor markers, which had a certain influence on the improvement of research design. Therefore, we will refer to the latest research in real-time in the later period and add corresponding research schemes to make up for design defects so as to continuously improve the research (45, 46).

In conclusion, miR-383 down-regulated in the placental tissue of patients with severe preeclampsia, while miR-16 up-regulated. miR-383 and miR-16 might participate in the process of occurrence, development and metastasis of severe preeclampsia, and could be used as potential biomarkers of placental tissue for its diagnosis and disease assessment of metastasis.

Authors' contributions

TL wrote the manuscript. BZ performed PCR. YH analyzed and interpreted the patients' data. JL and YL helped with statistical analysis. All authors read and approved the final manuscript.

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