Diagnostic Value of Serum miR-25-3p in Hypertensive Disorders in Pregnancy

Dexia Zhou (dexz0126@163.com)
Nantong Maternity and Child Health Hospital https://orcid.org/0000-0002-0140-3122
Bin Qu
Nantong Maternity and Child Health Hospital
Xuan Zhang
Nantong Maternity and Child Health Hospital

Research

Keywords: Hypertensive disorders in pregnancy, miR-25-3p, Diagnosis, Prognosis, Receiver operating characteristic curve, Adverse pregnancy outcomes, Mild preeclampsia, Severe preeclampsia

Posted Date: May 26th, 2021

DOI: https://doi.org/10.21203/rs.3.rs-524262/v1

Abstract

Objective: Hypertensive disorders in pregnancy (HDIP) are a variety of heterogeneous conditions. microRNA (miR)-25-3p is involved in HDIP diagnosis. This study explored the clinical effects of miR-25-3p on HDIP patients.

Methods: A total of 190 HDIP patients including 65 cases of gestation hypertension (GH), 67 cases of mild preeclampsia (mPE) and 58 cases of severe preeclampsia (sPEz), and 60 normal pregnant women were selected as the control. Serum miR-25-3p expression in HDIP patients and controls was detected. Diagnostic efficacy of miR-25-3p on HDIP was analyzed. HDIP patients were assigned into high/low miR-25-3p expression groups. sPEz patients assigned into high/low miR-25-3p expression group were followed up until delivery. Gestational weeks and pregnancy outcomes were recorded at delivery. The effect of miR-25-3p expression on pregnancy outcomes of sPE patients was analyzed.

Results: Compared with the controls, miR-25-3p expression in GH, mPE and sPEz patients was upregulated. miR-25-3p expression was highest in sPE patients, then in mPE patients and then in GH patients. Area under the curve of miR-25-3p for sPEz diagnosis was 0.946, with 87.93% sensitivity and 98.33% specificity, which were higher than those for GH and mPE diagnosis. In sPEz patients, systolic and diastolic blood pressure, 24-h urine protein, AST, ALT, GGT and Scr were increased, and PLT was decreased in high expression group. High miR-25-3p expression increased risks of adverse pregnancy outcome in sPEz patients.

Conclusions: High miR-25-3p expression helps HDIP diagnosis and is related to increased risks of adverse pregnancy outcomes in sPEz patients.

Introduction

Hypertensive disorders in pregnancy (HDIP) are a heterogeneous variety of conditions, which include gestational hypertension (GH) and preeclampsia (PE) [1]. HDIP are characterized by edema, proteinuria, and hypertension [2]. The nephrolithiasis history is related to a higher risk of HDIP, especially in the pregnant women who are with high body mass index (BMI) in early pregnancy [3]. At present, most of recommendations for HDIP treatment are based on observational studies and the opinion of experts, which lacks evidence from randomized controlled trials [4]. As HDIP increase the risk of the mortality and morbidity for perinatal and maternal, timely diagnosis and effective treatment are necessary to prevent the complications [3]. Therefore, it is of vital importance to find new biomarkers for HDIP diagnosis and treatment for patients with HDIP.

microRNAs (miRNAs) are the small non-coding RNAs, which have the effect of regulating gene expression at post-transcriptional level by promoting messenger RNA (mRNA) degradation or promoting mRNA translation [5]. The understanding of how miRNAs modulate gene expressions and influence various pathways that contribute to a variety of human pathologies has been significantly improving recently, while the effect of miRNAs on HDIP, such as PE is only beginning to emerge [6]. It has been reported that

the abnormal expression of a variety of miRNAs are related to the adverse pregnancy outcomes, and efforts are underway to explore biological effect of the placental miRNAs that can contribution to HDIP [7]. Recently, vascular endothelium that works as an endocrine organ, which can preserve the balance of homeostasis through responding to the metabolic status changes, has been recognized as a key factor in the initiation and the development of disorders in pregnancy [8]. PE is a pregnancy-specific disease with endothelial cell dysfunction, which is characterized by high blood pressure, proteinuria and edemas, influencing 3–5% of the pregnant women all over the world [9]. The expression of miR-25 is significantly upregulated in the placenta of PE patients [10]. Overexpression of lncRNA SNHG12 attenuates the endothelial injury in hypertensive mice by inhibiting the expression of miR-25-3p [11]. However, there is no domestic and foreign report at present on miR-25-3p expression and clinical research in serum of patients with HDIP. This study investigated the expression of miR-25-3p in serum of HDIP patients, and the clinical value of miR-25-3p in the diagnosis, severity evaluation and pregnancy outcome prediction of HDIP patients, so as to provide certain reference value for the diagnosis and prognosis of HDIP.

Materials And Methods

Ethics statement

The experiments were authorized by the academic ethics committee of Nantong Maternity and Child Health Hospital. All procedures were strictly implemented by the code of ethics. All the subjects involved were fully informed of the objective of the study and signed informed consent before sampling.

Study subjects

A total of 190 patients with HDIP who were treated in the outpatient department of Nantong Maternity and Child Health Hospital from January 2018 to December 2020 were selected, including 65 cases of GH (GH group, hypertension and no proteinuria after 20 weeks of gestation), 67 cases of mild preeclampsia (mPE group, systolic pressure ≥ 140 mmHg or diastolic pressure ≥ 90 mmHg, 24-h proteinuria ≥ 300 mg), and 58 cases of severe preeclampsia (sPEz group, systolic pressure ≥ 160 mmHg or diastolic pressure ≥ 100 mmHg, 24-h proteinuria ≥ 2g). The diagnosis of HDIP was based on the relevant diagnostic and grading standards of HDIP in the practice bulletin of American College of Obstetricians and Gynecologists (ACOG) in 2002 [12], and was confirmed by pathological examination. Another 60 cases of normal pregnant women were selected as the control group. Inclusion criteria were as follows: singleton pregnancy, age 20–40 years and no history of special medication. Exclusion criteria were as follows: hypertension history, pathological changes in the main organs such as liver, kidney and lung, autoimmune diseases, and pregnancy with anemia, diabetes, or heart disease.

Clinical pathological characteristics detection

The following clinical indicators were measured and recorded when the subjects were enrolled: age, gestational weeks, gestational times, pre-pregnancy BMI, systolic and diastolic blood pressure. The protein content in urine samples was determined using a urine protein quantitative test kit (Wanlei bio, Liaoning, China). The platelet count (PLT) was detected using an XS800i automatic hematology analyzer. The

contents of aspartate aminotransferase (AST), alanine aminotransferase (ALT), glutamyl transpeptidase (GGT) and serum creatinine (Scr) in blood samples were detected using a TCHG500 automatic biochemical analyzer.

Follow-up of adverse pregnancy outcomes

The enrolled patients were followed up until delivery. After delivery, the maternal and fetal outcomes were collected, and the gestational weeks and pregnancy outcomes were recorded. The adverse outcome of pregnancy was defined as: any of the circumstances of placenta abruption, heart dysfunction, acute renal injury, cerebrovascular accident, diffuse intravascular hemorrhage, premature delivery, postpartum hemorrhage, small for gestational age (SGA), asphyxia of newborn and stillbirth occurred during the follow-up or delivery. SGA was defined as that the 10th percentile of the birth weight was lower than the expected weight of the same gestational age. Preterm birth was defined as less than 37 weeks at delivery. Neonatal asphyxia was defined as Apgar score ≤ 7 at 1 min after birth.

Total RNA extraction and reverse transcription quantitative polymerase chain reaction (RT-qPCR) detection

The TRIzol reagent (Invitrogen, Carlsbad, CA, USA) was used to extract the total RNA. The PrimeScript RT reagent kit (Takara, Dalian, China) was used for reverse transcription of RNA into cDNA. The qPCR was performed on the ABI7900HT fast PCR real-time system (Applied Biosystems, Foster city, CA, USA) using SYBR® Premix Ex Taq™ II (Takara). The reaction conditions were pre-denaturation at 95℃ for 10 min and 40 cycles of denaturation at 95℃ for 10 s, annealing at 60℃ for 20 s and extending at 72℃ for 34 s. U6 was used as the internal parameter and the $2^{-\Delta\Delta Ct}$ method was used to analyze data [13]. The primer sequences (synthetized by Sangon Biotech Co., Ltd., Shanghai, China) are shown in Table 1.

Table 1
primer sequence

Gene	Forward 5′-3′	Reverse 5′-3′
miR-25-3p	GCACGCCGGCGACAGG	AGTGCAGGGTCCGAGGTATT
U6	ATTGGAACGATACAGAGAAGATT	GGAACGCTTCACGAATTTC

Statistical analysis

SPSS 21.0 software (IBM Corp. Armonk, NY, USA), GraphPad Prism 8 software (GraphPad Software Inc) and Medcalc® version 15.0 statistical software (Medcalc Software Ltd, Ostend, Belgium) were used for data analysis and mapping. The variable data were expressed as mean ± standard deviation or in counts. Non-paired *t* test or x^2 was used for comparison between two groups and one-way analysis of variance (ANOVA) was used for comparison among multi-groups. Tukey's multiple comparisons test was used for post hoc test. Receiver operating characteristic (ROC) curve was used for diagnostic analysis of miR-25-3p on HDIP. Kaplan-Meier method was used for analysis of the effect of miR-25-3p on pregnancy outcome on sPE patients. A value of $P < 0.05$ indicated statistical significance.

Results

Comparative analysis of clinical data between HDIP patients and healthy subjects

In this study, 190 patients with HDIP were assigned into 65 cases of GH, 67 cases of mPE and 58 cases of sPEz according to the relevant diagnostic and grading standards of HDIP, and 60 normal pregnant women were selected as controls. The clinical indexes of HDIP patients and the controls were compared and analyzed. There was no significant difference in age, pre-pregnancy BMI, gestational weeks and gestational times among the 4 groups. There were significant differences in systolic blood pressure, diastolic blood pressure, 24-h urine protein, PLT, AST, ALT, GGT and Scr among GH group, mPE group and sPEz group. There were also significant differences between mPE group and hypertention group, and between sPEz group and mPE group (all $P < 0.001$) (Table 2).

Table 2
Comparative analysis of clinical data between HDIP patients and controls

Parameters		Control group	GH group	mPE group	sPEz group
		(N = 60)	(N = 65)	(N = 67)	(N = 58)
Age (year)		29.15 ± 3.45	31.36 ± 3.83	30.54 ± 3.92	32.07 ± 3.16
Pre-pregnancy BMI (kg/m2)	< 25	36	35	38	33
	≥ 25	24	30	29	25
Gestational week	< 35	25	28	27	21
	≥ 35	35	37	40	37
Pregnancy time	< 1	40	46	50	44
	≥ 1	20	19	17	14
Systolic pressure (mmHg)		113.25 ± 10.36	136.17 ± 10.09^{a}	152.53 ± 10.42^{ab}	190.88 ± 10.56^{abc}
Diastolic pressure (mmHg)		76.13 ± 4.51	85.02 ± 4.66^{a}	93.49 ± 4.25^{ab}	108.87 ± 4.79^{abc}
24-h urine protein (g)		0.08 ± 0.03	0.15 ± 0.05^{a}	0.73 ± 0.21^{ab}	3.69 ± 0.83^{abc}
PLT (×10^{9}/L)		215.45 ± 19.67	206.60 ± 19.33^{a}	188.19 ± 18.35^{ab}	160.04 ± 18.22^{abc}
AST (U/L)		18.59 ± 7.15	29.91 ± 7.63^{a}	43.58 ± 8.77^{ab}	68.17 ± 8.39^{abc}
ALT (U/L)		11.09 ± 3.26	28.33 ± 3.85^{a}	34.65 ± 4.01^{ab}	47.71 ± 4.55^{abc}
GGT (U/L)		12.13 ± 5.54	24.96 ± 5.85^{a}	36.99 ± 6.46^{ab}	49.19 ± 6.93^{abc}
Scr (μmol/mL)		61.31 ± 6.77	75.58 ± 7.43^{a}	82.16 ± 7.94^{ab}	99.05 ± 9.15^{abc}

Note: GH (Gestational hypertension); mPE (mild preeclampsia); sPEz (severe preeclampsia); BMI (Body Mass Index); PLT (platelet count); AST (aspartate transaminase); ALT (Alanine transaminase); GGT (gamma-glutamyltransferase); Scr (serum creatinine). a presents $P < 0.05$ compared with the control group; b presents $P < 0.05$ compared with GH group; c presents $P < 0.05$ compared with mPE group.

miR-25-3p was highly expressed in serum of HDIP patients

The expression of miR-25-3p in serum of HDIP patients and controls was detected by RT-qPCR. Compared with the control group, the expression of miR-25-3p in patients of GH group, mPE group and sPEz group was significantly upregulated, and the expression of miR-25-3p in patients of mPE group was significantly higher than that of the GH group, and miR-25-3p expression in patients of sPEz group was significantly higher than that of mPE group (all $P < 0.001$) (Fig. 1).

miR-25-3p had high clinical diagnostic efficiency in patients with sPEz

In order to further study the clinical diagnostic significance of serum miR-25-3p expression in HDIP, the diagnostic efficacy of miR-25-3p on HDIP was analyzed by ROC curve. The area under the curve (AUC) of miR-25-3p in serum for the diagnosis of GH was 0.770, the specificity was 98.33%, and the sensitivity was 49.23% (Fig. 2A); AUC for the diagnosis of mPE was 0.848, the specificity was 93.33%, and the sensitivity was 74.63% (Fig. 2B); and AUC for the diagnosis of sPEz was 0.946, the specificity was 98.33%, and the sensitivity was 87.93% (Fig. 2C). The results indicated that miR-25-3p had highest clinical diagnostic efficiency in patients with sPEz.

Correlation analysis of miR-25-3p expression and clinical indicators of HDIP patients

According to the median value of miR-25-3p expression, the patients with HDIP, mPE and sPEz were assigned into low expression group and high expression group, and then the correlation with the clinical indicators of HDIP patients was analyzed. In patients with HDIP, mPE and sPEz, there were no significant differences in age, pre-pregnancy BMI, gestational weeks and gestational times between the high expression group and the low expression group. In addition, AST and GGT were significantly increased in GH patients with high miR-25-3p expression group compared with those of the low miR-25-3p expression group (all $P < 0.05$), but there was no significant difference in other clinical indicators. In patients with mPE, systolic blood pressure, AST and ALT were significantly higher in the miR-25-3p high expression group than those of the low expression group (all $P < 0.05$), but there was no significant difference in other clinical indicators. In the patients with sPEz, systolic pressure, diastolic pressure, 24-h urine protein content, AST, ALT, GGT and Scr were significantly increased, and PLT was significantly decreased in the high miR-25-3p expression group than those of the low miR-25-3p expression group (all $P < 0.05$) (Table 3).

Table 3
Correlation between serum miR-25-3p expression and clinical indicators in patients with HDIP

Parameters		GH (N = 65)		mPE (N = 67)		sPEz (N = 58)	
		Low expression	High expression	Low expression	High expression	Low expression	High expression
		(N = 32)	(N = 33)	(N = 33)	(N = 34)	(N = 29)	(N = 29)
Age (year)		32.11 ± 3.62	30.45 ± 3.55	31.23 ± 3.64	29.56 ± 3.71	33.05 ± 3.46	31.70 ± 3.57
Pre-pregnancy BMI (kg/m^2)	< 25	17	18	20	18	15	18
	≥ 25	15	15	13	16	14	11
Gestational week	< 35	15	13	15	12	11	10
	≥ 35	17	20	18	22	18	19
Pregnancy times	< 1	20	26	22	28	21	23
	≥ 1	12	7	11	6	8	6
Systolic pressure (mmHg)		136.20 ± 9.39	136.14 ± 10.87	149.59 ± 9.96	155.38 ± 10.19[b]	186.25 ± 9.51.	195.51 ± 9.60[c]
Diastolic pressure (mmHg)		84.42 ± 5.44	85.60 ± 3.75	92.96 ± 3.95	94.00 ± 4.52	106.64 ± 3.82	111.10 ± 4.67[c]
24-h urine protein (g)		0.15 ± 0.05	0.15 ± 0.05	0.72 ± 0.21	0.74 ± 0.21	3.32 ± 0.75	4.06 ± 0.75[c]
PLT ($1\times10^9/L$)		208.02 ± 19.76	205.23 ± 19.11	190.60 ± 18.15	185.85 ± 18.51	168.21 ± 17.00	151.87 ± 15.77[c]
AST (U/L)		27.98 ± 7.98	31.78 ± 6.89[a]	41.17 ± 8.57	45.92 ± 8.44[b]	64.82 ± 7.61	71.52 ± 7.89[c]
ALT (U/L)		28.18 ± 3.78	28.47 ± 3.97	33.32 ± 3.69	35.94 ± 3.93[b]	46.30 ± 4.14	49.12 ± 4.57[c]

Note: GH (Gestational hypertension); mPE (mild preeclampsia); sPEz (severe preeclampsia); BMI (Body Mass Index); PLT (platelet count); AST (aspartate transaminase); ALT (Alanine transaminase); GGT (gamma-glutamyltransferase); Scr (serum creatinine). a presents in GH patients, miR-25-3p high expression group compared with low expression group $P < 0.05$; b presents in mPE patients, miR-25-3p high expression group compared with low expression group $P < 0.05$; c presents in sPEz patients, miR-25-3p high expression group compared with low expression group $P < 0.05$.

Parameters	GH (N = 65)		mPE (N = 67)		sPEz (N = 58)	
	Low expression	High expression	Low expression	High expression	Low expression	High expression
	(N = 32)	(N = 33)	(N = 33)	(N = 34)	(N = 29)	(N = 29)
GGT (U/L)	23.32 ± 5.86	26.55 ± 5.46[a]	36.54 ± 6.21	37.43 ± 6.76	46.16 ± 6.89	52.22 ± 5.59[c]
Scr (μmol/mL)	76.00 ± 7.40	75.17 ± 7.55	82.00 ± 8.48	82.31 ± 7.50	95.94 ± 7.51	102.16 ± 9.69[c]

Note: GH (Gestational hypertension); mPE (mild preeclampsia); sPEz (severe preeclampsia); BMI (Body Mass Index); PLT (platelet count); AST (aspartate transaminase); ALT (Alanine transaminase); GGT (gamma-glutamyltransferase); Scr (serum creatinine). a presents in GH patients, miR-25-3p high expression group compared with low expression group $P < 0.05$; b presents in mPE patients, miR-25-3p high expression group compared with low expression group $P < 0.05$; c presents in sPEz patients, miR-25-3p high expression group compared with low expression group $P < 0.05$.

High miR-25-3p expression increased the risk of adverse pregnancy outcomes in patients with sPE

The above results showed that miR-25-3p was highly expressed in the serum of patients with sPE and had high diagnostic efficiency, and was closely related to the clinical indicators of patients. In order to further explore the value of miR-25-3p in the prognosis of sPE patients, the sPE patients were assigned into low expression group and high expression group according to the median value of miR-25-3p expression. The two groups were followed up till delivery, and the gestational weeks and pregnancy outcomes were recorded. The incidence of adverse pregnancy outcomes between the two groups was compared. There were 8 cases of adverse pregnancy outcomes in the low expression group, and the incidence of adverse pregnancy outcomes was 27.6%. There were 17 cases of adverse pregnancy outcomes in the high expression group, and the incidence of adverse pregnancy outcomes was 58.6%, which was significantly higher than that in the low expression group ($x^2 = 5.70$, $P < 0.05$). In addition, Kaplan-Meier analysis showed that the curve of miR-25-3p high expression group shifted to the left ($P < 0.05$) (Fig. 3), indicating that at the same gestational age, the cumulative incidence of adverse pregnancy outcomes was higher in the high expression group. The results indicated that high expression of miR-25-3p was associated with poor pregnancy outcome.

Discussion

HDIP have has been affecting over 10% of the pregnant women all over the world, which accounts for a remarkable proportion of the mortality and morbidity of the perinatal and maternal [1]. Evidence has shown that miR-25-3p play an essential role in HDIP [10]. This study found that high expression of miR-25-3p could assist the diagnosis of HDIP.

Some miRNAs have been reported to be potential early biomarkers for HDIP [14]. However, the expression and the diagnostic value of miR-25-3p in HDIP were still unclear. Our results showed that the expression of miR-25-3p in GH, mPE, and sPEz patients was upregulated significantly compared with the normal pregnant women, and the expression of miR-25-3p in mPE patients was significantly higher than that of the GH patients, and that in sPE patients was significantly higher than that of mPE patients. It is consistent with that compared with the normal pregnant women, miR-25 was significantly overexpressed in PE patients [10]. Collectively, miR-25-3p was highly expressed in serum of HDIP patients and high expression of miR-25-3p could help diagnose the severity of HDIP.

In order to further study miR-25-3p expression in the clinical diagnosis of HDIP, we analyzed the miR-25-3p expression using ROC curve. Our results demonstrated that the AUC of miR-25-3p for the diagnosis of GH was 0.770, the specificity was 98.33, and the sensitivity was 49.23%. The AUC of miR-25-3p for the diagnosis of mPE was 0.848, the specificity was 93.33%, and the sensitivity was 74.63%. The AUC of miR-25-3p for the diagnosis of sPEz was 0.946, the specificity was 98.33%, and the sensitivity was 87.93%. In summary, high expression of miR-25-3p in serum of sPE patients had a highest clinical diagnostic efficiency. Overall, our results showed for the first time the diagnostic value of serum miR-25-3p in HDIP.

PE is characterized by high blood pressure and proteinuria [15]. PLT is a severity marker for PE [16]. AST and ALT levels are important indexes in PE [17]. Our results demonstrated that there were significant differences in systolic and diastolic pressure, 24-h urine protein, PLT, AST, ALT, GGT and Scr between GH, mPE and sPEz patients, and normal pregnant women. Consistently, elevated PLT, and systolic and diastolic pressure are the factors to diagnose HDIP [18, 19]. In conclusion, GH, mPE and sPEz patients, and normal pregnant women showed different clinical indexes, which may be useful to distinguish the normal population from HDIP patients. Furthermore, we analyzed the correlation between miR-25-3p expression and clinical indexes in patients with HDIP, and found that miR-25-3p expression was positively correlated with AST and GGT in GH patients, positively correlated with systolic and diastolic pressure, AST, and ALT in mPE patients, positively correlated with systolic, diastolic pressure, 24-h urine protein content, AST, ALT, GGT and Scr, and negatively correlated with PLT in sPEz patients. In recurrent pregnancy loss, blood urea nitrogen levels decreased depending on the miR-25 allele and creatinine levels were also associated with the genotypes of the miR-25 alleles [20]. However, there is no domestic and foreign report at present on the clinical diagnostic efficiency of miR-25-3p expression and its correlation with clinical indexes in serum of patients with HDIP. These results indicated that miR-25-3p had close correlations with the clinical parameters of HDIP.

Moreover, many circulating miRs are closely associated with adverse pregnancy outcomes [21]. We analyzed the association of miR-25-3p level with adverse pregnancy outcomes. Our results demonstrated that there were 8 cases of adverse pregnancy outcomes in the low expression patients, and the incidence of adverse pregnancy outcomes was 27.6%; there were 17 cases of adverse pregnancy outcomes in the high expression patients, and the incidence of adverse pregnancy outcomes was 58.6%, indicating that the adverse outcomes of the pregnant women with high expression of miR-25-3p were higher than those with low expressions. The overexpression of miR-25 is reported to be associated with cardiac insufficiency [22].

However, there are little studies on the relationship between miR-25-3p level and the prognosis of HDIP. Our study initially revealed that miR-25-3p was associated with adverse pregnancy outcomes in HDIP patients.

In summary, this study supported that miR-25-3p was highly expressed in serum of patients with HDIP. Overexpression of miR-25-3p could help diagnose the severity of HDIP, and was related to the increased risk of adverse pregnancy outcome in patients with sPEz, which provided a new reference for HDIP diagnosis, severity evaluation and pregnancy outcome prediction. However, the number of cases and events included in this study is limited. It is necessary to further expand the sample size and carry out a multi-center study to further clarify the diagnostic and prognostic evaluation ability of miR-25-3p for HDIP. In order to increase the reliability of the results, further study is needed to carry out the multi-center prospective study to explore the molecular regulation mechanism of miR-25-3p in the development of HDIP.

Declarations

Acknowledgements

Not applicable.

Funding

Not applicable.

Conflict of Interests

The authors declare that they have no conflicts of interest.

Data Availability Statement

All the data generated or analyzed during this study are included in this published article.

Ethics approval and consent to participate

The experiments were authorized by the academic ethics committee of Nantong Maternity and Child Health Hospital. All procedures were strictly implemented by the code of ethics. All the subjects involved were fully informed of the objective of the study and signed informed consent before sampling.

Consent for publication

Not applicable.

Authors' Contributions

DXZ is the guarantor of integrity of the entire study; DXZ contributed to the study design, definition of intellectual content, literature research, clinical studies, experimental studies, data acquisition, data analysis, statistical analysis, manuscript preparation, manuscript editing, and manuscript review; BQ contributed to the study concepts and manuscript review; XZ contributed to the data acquisition and data analysis; All authors read and approved the final manuscript.

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Figures

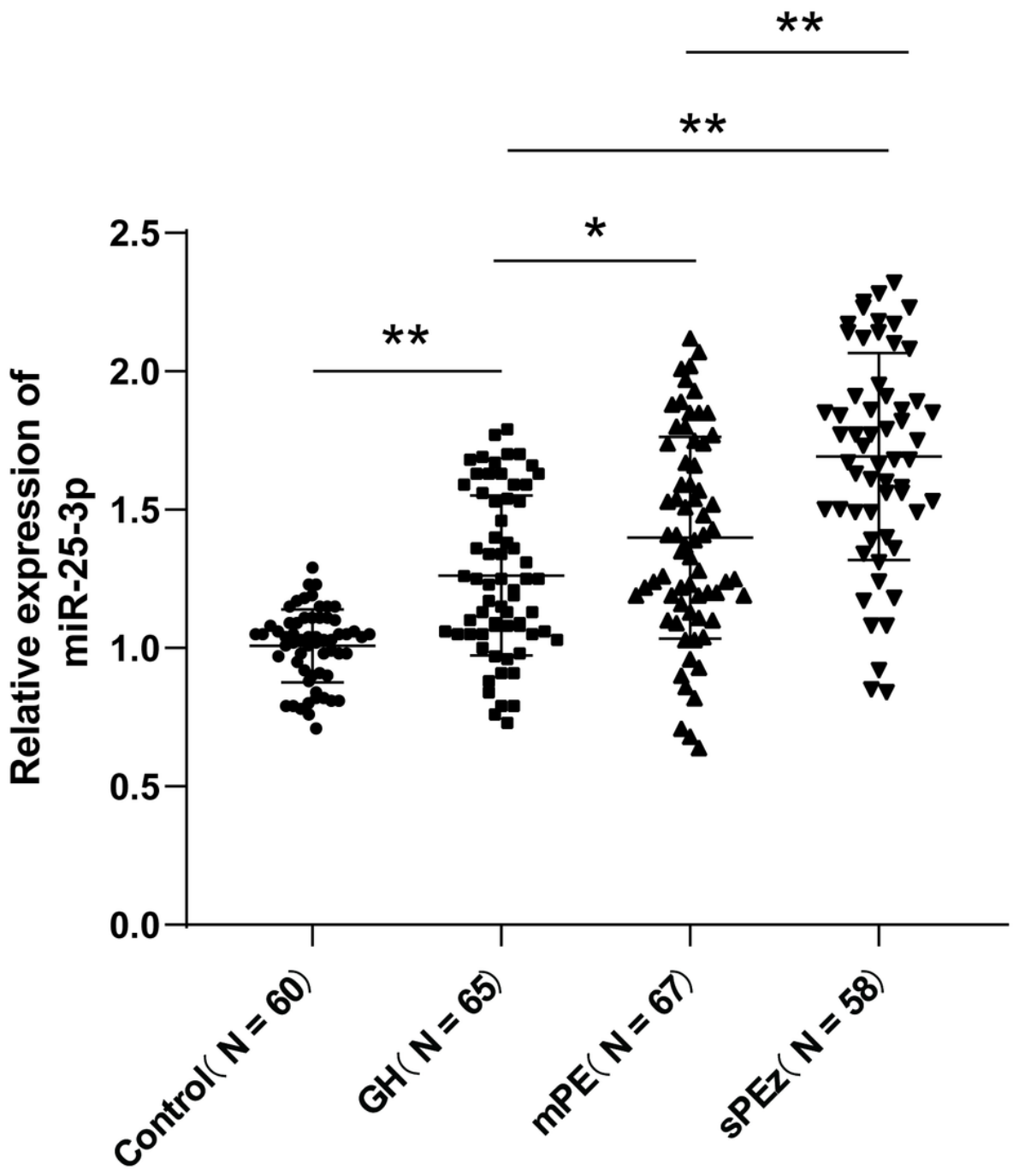

Figure 1

miR-25-3p was highly expressed in serum of HDIP patients. The expression of miR-25-3p in serum of HDIP patients and controls was detected by RT-qPCR. The data were expressed as mean ± standard deviation. One-way ANOVA was used for comparison among multi-groups. Tukey's multiple comparisons test was used for post hoc test. *$p < 0.05$, ** $p < 0.01$.

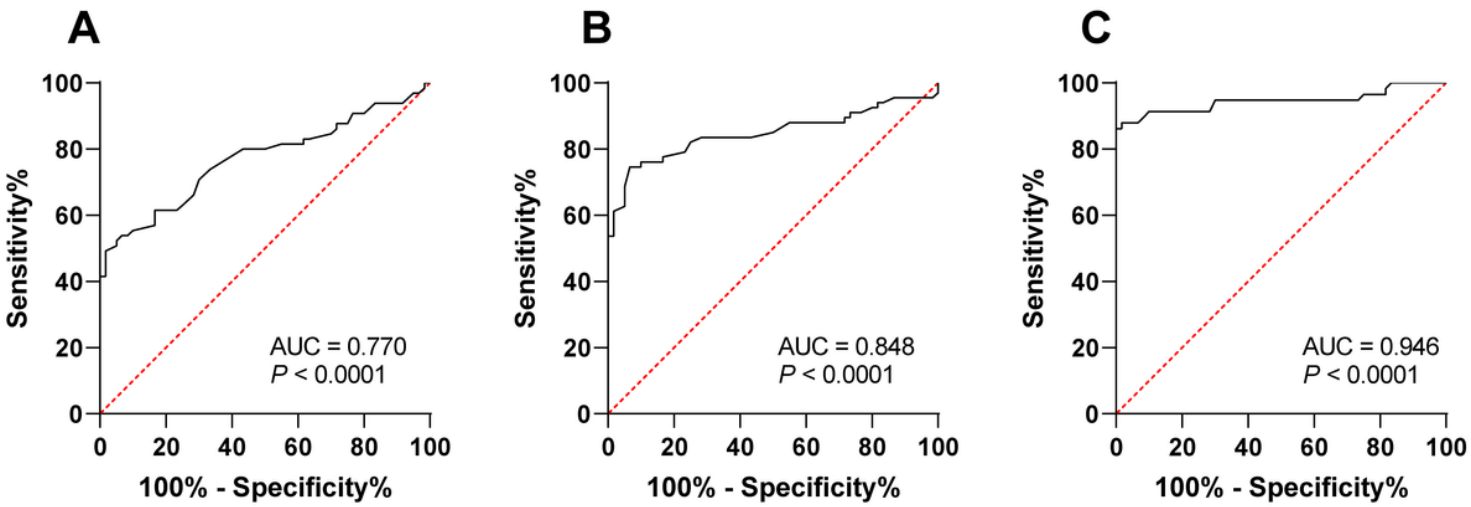

Figure 2

Diagnostic efficacy of miR-25-3p in patients with HDIP. The diagnostic efficacy of miR-25-3p in patients with (A) GH; (B) mPE; and (C) sPEz was analyzed using ROC curve.

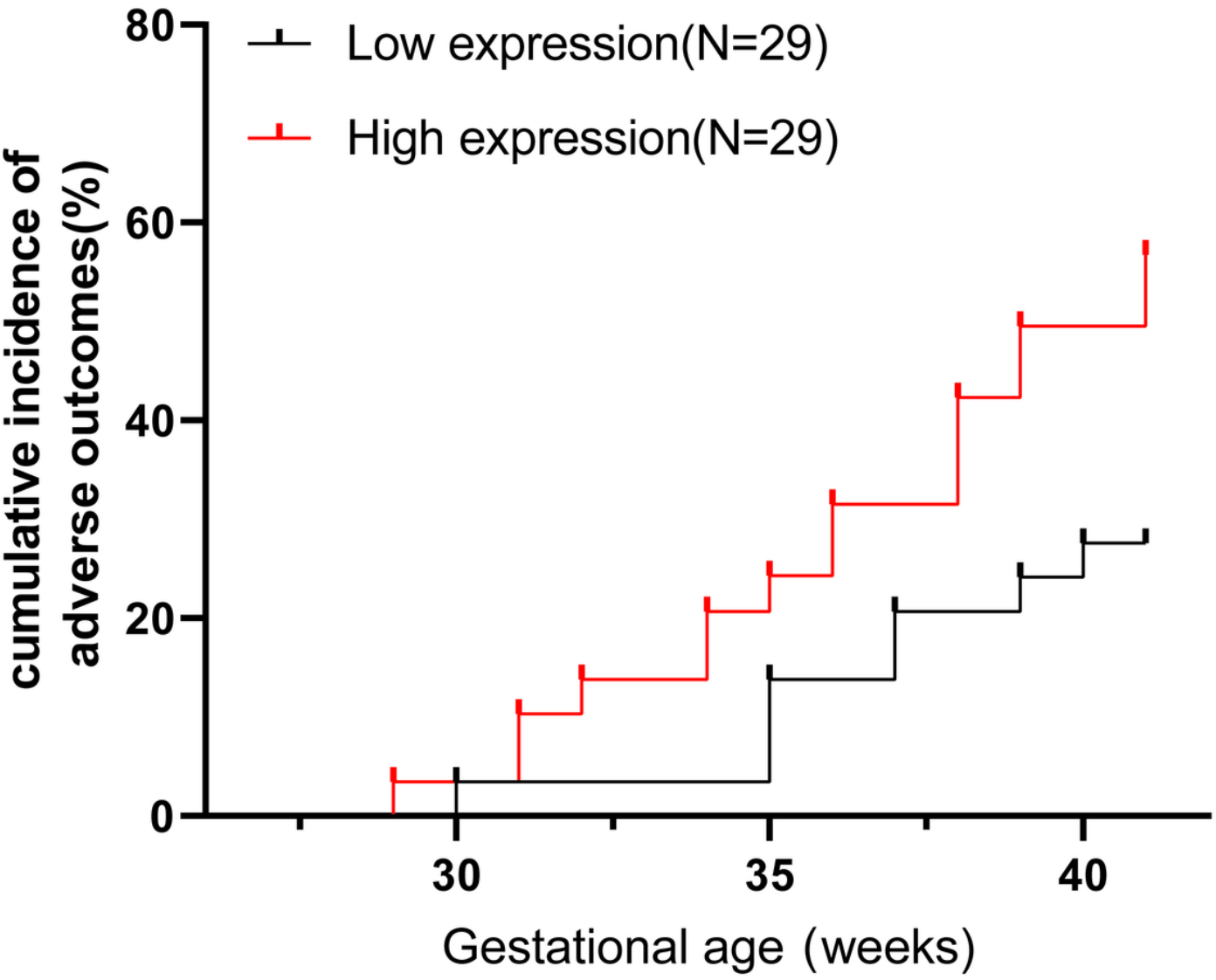

Figure 3

High expression of miR-25-3p increased the risk of adverse pregnancy outcome in patients with sPEz. The effect of miR-25-3p on pregnancy outcome in patients with sPEz was analyzed using Kaplan-Meier method.