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RESEARCH ARTICLE

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A potential disease monitoring and prognostic biomarker in cervical cancer patients: The clinical application of circular RNA_0018289

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Abstract

Objective: This study aimed to investigate the tumor circular RNA_0018289 (circ_0018289) expression and its correlation with clinical characteristics as well as survival profiles in cervical cancer patients.

Methods: A hundred and ninety-two cervical cancer patients who received surgical resection were recruited in this prospective study. Tumor tissue and paired adjacent tissue were obtained during the surgery, in which circ_0018289 expression was detected by reverse transcription quantitative polymerase chain reaction. Disease-free survival (DFS) and overall survival (OS) were recorded.

Results: Circ_0018289 expression was upregulated in tumor tissue compared with paired adjacent tissue ($P < .001$), and receiver operative characteristic curve disclosed its good value for separating tumor tissue from adjacent tissue with an area under curve of 0.907 (95% CI: 0.879-0.935). Additionally, tumor circ_0018289 expression was positively associated with tumor size ($P = .009$), lymph node metastasis ($P = .005$) and Federation International of Gynecology and Obstetrics stage ($P = .005$). The DFS ($P = .005$) and OS ($P = .015$) were both worse in patients with circ_0018289 high expression compared to patients with circ_0018289 low expression. Meanwhile, in patients with circ_0018289 high expression, DFS and OS were the longest in patients with high+ expression followed by patients with high++ expression, and the shortest in patients with high+++ expression. Moreover, circ_0018289 high expression could independently predict worse DFS in the total cervical cancer patients ($P = .042$).

Conclusion: Circ_0018289 could serve as a potential disease monitoring and prognostic biomarker in cervical cancer patients.

KEYWORDS

cervical cancer, circ_0018289, clinical characteristics, survival, tumor tissue

Jing He and Xin Lv contributed equally to this work.

1 | INTRODUCTION

Cervical cancer is ranked as the fourth most fatal cancer in females and presents with an increasing premature mortality rate in Asian countries due to the lack of adequate and organized screening program.[1-4] Prolonging survival of patients is the mainstay of treatment goal of almost all cancers as well as for the management of cervical cancer. Nowadays, the therapies available for cervical cancer patients include hysterectomy, chemoradiation, and targeted therapy. However, despite that screening, diagnosis and novel treatments have largely progressed, the survival profile of cervical cancer patients is still unsatisfactory especially for the metastatic patients and relapsed patients.[5,6] Therefore, detecting useful biomarkers that may assist in disease management is urgent for cervical cancer patients.

Circular RNAs (circRNAs) are a class of endogenous RNAs with almost no coding ability. They are categorized by having a closed-loop structure, which allows a more stable expression.[7] Although circRNAs are ignored at their discovery, increasing functions of circRNAs in human diseases are uncovered in recent years.[8] Interestingly, a previous study conducted by our collaborate institution illustrates a circRNA that might participate in the pathogenesis of cervical cancer, which is circ_0018289.[9] In their study, circ_0018289 is identified by microarray to be upregulated in tumor tissue compared with adjacent non-tumor tissue; then, reverse transcription-quantitative polymerase chain reaction (RT-qPCR) validates that it is increased in both tumor tissue and cancer cells; moreover, the further experiment shows that circ_0018289 knockdown inhibits tumor-like behaviors of cancer cells in vitro as well as represses tumor growth in vivo.[9] Hence, we speculated that circ_0018289 may have the potential to serve as a disease monitoring and prognostic biomarker for cervical cancer in the clinical setting. Nonetheless, to the best knowledge of ours, no study has been done to validate this presumption.

Thus, this study aimed to assess the tumor circ_0018289 expression and its correlation with clinical characteristics as well as survival profiles in cervical cancer patients.

2 | MATERIALS AND METHODS

2.1 | Patients

A total of 192 cervical cancer patients who received surgical resection in our hospital between July 2015 and June 2019 were recruited in this prospective study. The inclusion criteria were (a) diagnosed as primary cervical cancer; (b) Federation International of Gynecology and Obstetrics (FIGO) stage I-IIA; (c) ready for surgical resection without neoadjuvant therapy; and (d) aged more than 18 years. The exclusion criteria were (a) relapsed cervical cancer; (b) severe dysfunction of hemogram, liver, or kidney; (c) complicated with other malignancies or history of other malignancies; (d) history of gynecologic surgery; (e) pregnant or lactating women; and (f) pregnant or lactating women. This study was approved by the Ethics Committee of our hospital. Written informed consents were collected from all patients.

2.2 | Clinical data and tissue sample collection

Before surgery, the key clinical data of enrolled patients were documented, including age, human papillomavirus (HPV) status, histological type, pathological grade, tumor size, lymph node status, and FIGO stage. During the surgery, the removed tumor tissue as well as adjacent tissue were snap-frozen in liquid nitrogen and preserved in an ultra-cold storage freezer for further detection of circ_0018289 expression by the RT-qPCR assay.

2.3 | RT-qPCR assay

First, the total RNA was extracted from tissues by the RNeasy Protect Mini Kit (Qiagen), which was then assessed by a spectrophotometer for detecting the purity and concentration. Afterwards, the linear RNA was digested by RNase R (Epicentre). Secondly, a PrimeScript™ RT reagent Kit (Perfect Real Time) (Takara) was used to reversely transcribe the RNA, and PCR was subsequently performed using the TB Green™ Fast qPCR Mix (Tayaba). In addition, the qPCR parameters were as follows: 95°C for 30 seconds, then 95°C for 5 seconds, 61°C for 15 seconds up to a total of 40 cycles; the melting curve parameters were as follows: 95°C for 5 seconds, 60°C for 1 minutes, 95°C (0.1°C/s), and 50°C for 30 seconds. Lastly, the relative expression of circ_0018289 was calculated in the formula $2^{-\Delta\Delta Ct}$ using glyceraldehyde-phosphate dehydrogenase (GAPDH) as the internal reference.[10] Primers were designed referring to the previous study[9]: circ_0018289, forward, 5′-TCACCAACCTTTGCCCTTCACACCT-3′, reverse, 5′-AAGACTTACGTCTGTGTGCGTTGT-3′; GAPDH, forward, 5′-TCGACAGTCAGCCGCATCTTCTTT-3′, reverse, 5′-ACCAAATCCGTTGACTCCGACCTT-3′. For statistical analysis, the tumor circ_0018289 expression was categorized as tumor circ_0018289 low expression (in the 0 to 50th percentile (0-2.675)) and tumor circ_0018289 high expression (in the 50th to 100th percentile) using the median of tumor circ_0018289 expression as cutoff value. Further, also using circ_0018289 median expression as cutoff value, the tumor circ_0018289 high expression was categorized as tumor circ_0018289 high+ expression (in the 50th to 75th percentile (2.675-4.575)), tumor circ_0018289 high++ expression (in the 75th to 90th percentile (4.575-6.327)), and tumor circ_0018289 high+++ expression (in the 90th to 100th percentile (6.327-7.054)).

2.4 | Follow-up

After surgery, all patients were regularly followed up by clinic visit or phone call until June 30, 2019. The median follow-up duration was 24.0 months, ranging from 1.0 to 48.0 months. The survival

TABLE 1 Clinical characteristics of cervical cancer patients

Items	Cervical cancer patients (N = 192)
Age (y), mean ± SD	48.3 ± 10.1
<45 y, No. (%)	82 (42.7)
≥45 y, No. (%)	110 (57.3)
HPV status, No. (%)	
Negative	39 (20.3)
Positive	153 (79.7)
Histological type, No. (%)	
Adenosquamous carcinoma	10 (5.2)
Adenocarcinoma	40 (20.8)
Squamous carcinoma	142 (74.0)
Pathological grade, No. (%)	
G1	55 (28.7)
G2	79 (41.1)
G3	58 (30.2)
Tumor size, No. (%)	
<4 cm	108 (56.2)
≥4 cm	84 (43.8)
Lymph node metastasis, No. (%)	
No	157 (81.8)
Yes	35 (18.2)
FIGO stage, No. (%)	
I	119 (62.0)
IIA	73 (38.0)

Abbreviations: FIGO, International Federation of Gynecology and Obstetrics; HPV, human papillomavirus; SD, standard deviation.

status during the follow-up was documented in detail for assessment of disease-free survival (DFS) and overall survival (OS). DFS was calculated from the date of surgery to the date of an event occurred, which was defined as disease recurrence, disease progression, or death (whichever occurred first). Patients who did not experience a DFS event were censored on their last date of disease assessment. OS was calculated from the date of surgery to the date of death, and patients who were thought to be alive at the time of final analysis were censored on the last date of contact. In addition, the patients who were lost to follow-up were not included in this study.

2.5 | Statistical analysis

Statistical analysis was performed using SPSS 24.0 (IBM, USA), and figures were plotted using the GraphPad Prism 7.02 (GraphPad Software Inc, USA). Data were described as mean and standard deviation (SD), median and interquartile range (IQR) or count (percentage), and comparison was determined by the chi-square test, Wilcoxon rank sum test, or Wilcoxon signed-rank sum test. Receiver operating characteristic (ROC) curve analysis and the derived area under curve (AUC) were used for assessing the performance of circ_0018289 in distinguishing tumor and adjacent tissue. DFS and OS were displayed using the Kaplan-Meier curves, and comparisons of DFS and OS between or among groups were determined by the log-rank test. Univariate cox's regression and backward stepwise multivariate cox's regression were performed to analyze factors predicting DFS and OS P value < .05 was considered statistically significant.

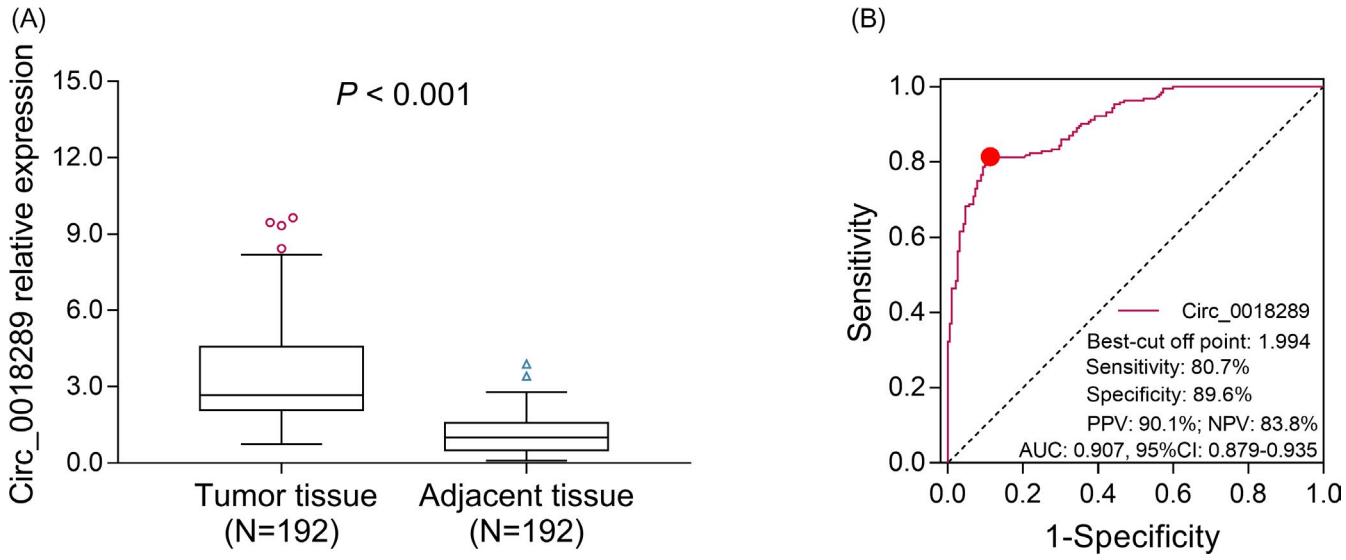

FIGURE 1 Difference of circ_0018289 expression in tumor tissue and paired adjacent tissue. The relative expression of circ_0018289 in tumor tissue and paired adjacent tissue (A), and ROC curve presenting the value of circ_0018289 for differentiating tumor tissue from paired adjacent tissue (B). AUC, area under curve; CI, confidence interval; Circ, circular RNA; NPV, negative predictive value; PPV, positive predictive value; ROC, receiver operating characteristic

TABLE 2 Comparison of clinical characteristics between circ_0018289 high expression patients and circ_0018289 low expression patients

Items	Circ_0018289 expression		*P* value
	High (n = 96)	Low (n = 96)	
Age (y), No. (%)			.559
<45 y	43 (44.8)	39 (40.6)	
≥45 y	53 (55.2)	57 (59.4)	
HPV status, No. (%)			.590
Negative	21 (21.9)	18 (18.8)	
Positive	75 (78.1)	78 (81.2)	
Histological type, No. (%)			.421
Adenosquamous carcinoma	7 (7.3)	3 (3.1)	
Adenocarcinoma	19 (19.8)	21 (21.9)	
Squamous carcinoma	70 (72.9)	72 (75.0)	
Pathological grade, No. (%)			.218
G1	27 (28.1)	28 (29.1)	
G2	34 (35.4)	45 (46.9)	
G3	35 (36.5)	23 (24.0)	
Tumor size, No. (%)			.009
<4 cm	45 (46.9)	63 (65.6)	
≥4 cm	51 (53.1)	33 (34.4)	
Lymph node metastasis, No. (%)			.005
No	71 (74.0)	86 (89.6)	
Yes	25 (26.0)	10 (10.4)	
FIGO stage, No. (%)			.005
I	50 (52.1)	69 (71.9)	
IIA	46 (47.9)	27 (28.1)	

Note: Comparison was determined by chi-square test or Wilcoxon rank sum test.

Abbreviations: FIGO, International Federation of Gynecology and Obstetrics; HPV, human papillomavirus.

3 | RESULTS

3.1 | The cervical cancer patients' clinical characteristics

The cervical cancer patients in our study had an average age of 48.3 ± 10.1 years, and there were 82 (42.7%) patients who were <45 years and 110 (57.3%) patients who were ≥45 years (Table 1). In addition, the number of patients with negative HPV status and patients with positive HPV status was 39 (20.3%) and 153 (79.7%), respectively. And there were 10 (5.2%), 40 (20.8%), and 142 (74.0%) patients who had histological type of adenosquamous carcinoma, adenocarcinoma, and squamous carcinoma, respectively. The number of patients at pathological grade of G1, G2, and G3 was 55 (28.7%), 79 (41.1%), and 58 (30.2%), respectively. Patients who had a tumor size <4 cm as well as patients with a tumor size ≥4 cm were 108 (56.2%) and 84 (43.8%), respectively. A total of 157 (81.8%) patients had no lymph node metastasis, and the other 35 (18.2%) patients had lymph node metastasis. Besides, the numbers of patients in FIGO stage I and FIGO stage IIA were 119 (62.0%) and 73 (38.0%), respectively.

3.2 | The expression of circ_0018289 in tumor tissue and paired adjacent tissue

In cervical cancer patients, circ_0018289 was upregulated in tumor tissue compared with paired adjacent tissue ($P < .001$) (Figure 1A). Additionally, the expression (presented as Ct value) of the reference gene GAPDH in tumor tissue and paired adjacent tissue was displayed in Supplementary Table S1. Furthermore, ROC curve analysis revealed that circ_0018289 could clearly separate tumor tissue from adjacent tissue with an AUC of 0.907 (95% CI: 0.879-0.935) (Figure 1B). Besides, the best cutoff value, sensitivity, specificity, negative predictive value (NPV), and positive predictive value (PPV) were 1.994, 80.7%, 89.6%, 90.1%, and 83.8%, respectively.

3.3 | Correlation of circ_0018289 expression in tumor tissue with patients' clinical characteristics

As for the association of tumor circ_0018289 with clinical characteristics in cervical cancer patients, it was found that tumor circ_0018289 was positively associated with tumor size ($P = .009$), lymph node metastasis ($P = .005$), and FIGO stage ($P = .005$) (Table 2).

3.4 | Correlation of circ_0018289 expression in tumor tissue with patients' DFS and OS

In terms of patients' survival profiles, the DFS was less favorable in patients with circ_0018289 high expression compared to patients with circ_0018289 low expression ($P = .005$) (Figure 2A). Additionally, the DFS was the worst in patients with circ_0018289 high+++ expression, followed by patients with circ_0018289 high++ expression and circ_0018289 high+ expression, and the best in patients with circ_0018289 low expression ($P = .002$) (Figure 2B). As to OS, it was worse in patients with circ_0018289 high expression than that in patients with circ_0018289 low expression ($P = .015$) (Figure 3A). Besides, the OS was the shortest in patients with circ_0018289 high+++ expression, followed by patients with circ_0018289 high++ expression and circ_0018289 high+ expression, and the longest in patients with circ_0018289 low expression ($P = .019$) (Figure 3B).

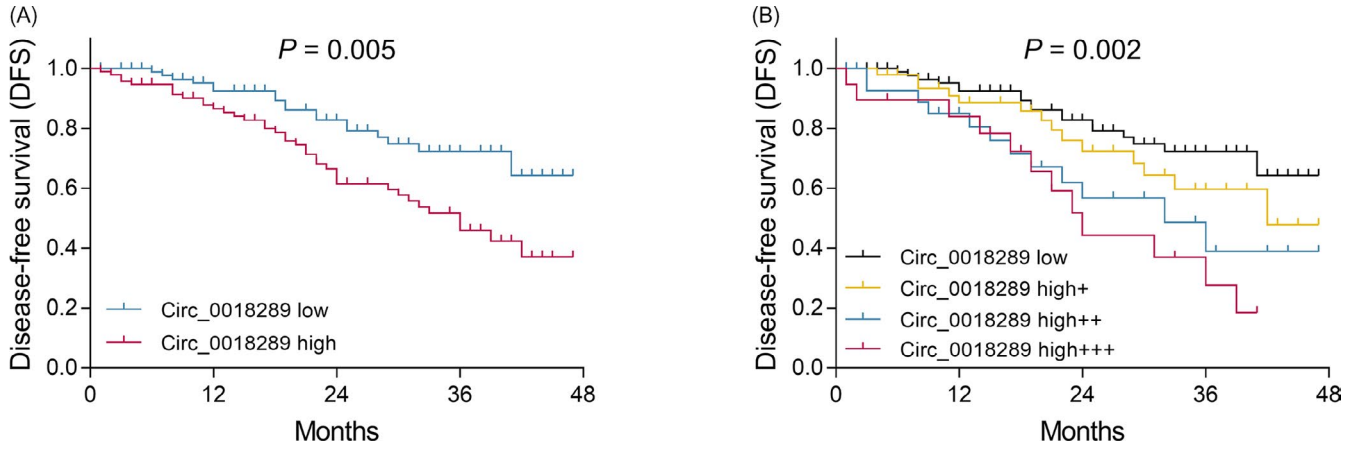

FIGURE 2 DFS in patients with different tumor circ_0018289 expressions. The DFS between patients with circ_0018289 high expression and patients with circ_0018289 low expression (A), and the DFS among patients with circ_0018289 low, high+, high++, as well as high+++ expression (B). circ, circular RNA; DFS, disease-free survival

3.5 | Analyses of prognostic factors

Predictive factors for DFS and OS were evaluated by Cox's regression analyses. The univariate Cox's regression analysis disclosed that circ_0018289 high expression could predict shorter DFS ($P = .006$); meanwhile, higher pathological grade ($P < .001$), tumor size ≥4 cm ($P = .011$), lymph node metastasis ($P = .005$), and higher FIGO stage ($P < .001$) also predicted pejorative DFS (Table 3). Then, the multivariate Cox's regression using backward stepwise method was conducted, which revealed that circ_0018289 high expression could independently predict worse DFS ($P = .042$); meanwhile, higher pathological grade ($P < .001$) and higher FIGO stage ($P = .023$) were also independent predictive factors for worse DFS. With regard to OS, the univariate Cox's regression analysis displayed that circ_0018289 ($P = .018$) was a predictive factor for shorter OS, and higher pathological grade ($P < .001$), tumor size ≥4 cm ($P = .002$), lymph node metastasis ($P = .008$), and higher FIGO stage ($P < .001$) were also predictors for shorter OS (Table 4). Moreover, the backward stepwise multivariate Cox's regression analysis showed that higher pathological grade ($P = .001$), tumor size ≥4 cm ($P = .013$), and higher FIGO stage ($P = .019$) were independent predictors for worse OS. Furthermore, the ROC curve analysis revealed that the AUC of tumor circ_0018289 for predicting relapse or death within 48 months was 0.671 (95% CI: 0.585-0.757), with the sensitivity, specificity, PPV, and NPV at the best cutoff value of 82.4%, 44.6%, 51.0%, and 78.3%, respectively (Figure 4A). In addition, the AUC of tumor circ_0018289 in predicting death within 48 months was 0.683 (95% CI: 0.590-0.776), and the sensitivity, specificity, PPV, and NPV at the best cutoff value were 78.3%, 50.0%, 37.7%, and 85.6%, respectively (Figure 4B).

3.6 | Correlation of circ_0018289 expression grade with clinical characteristics

The tumor circ_0018289 expression grade was positively correlated with pathological grade ($P = .038$), tumor size ($P = .011$), lymph node

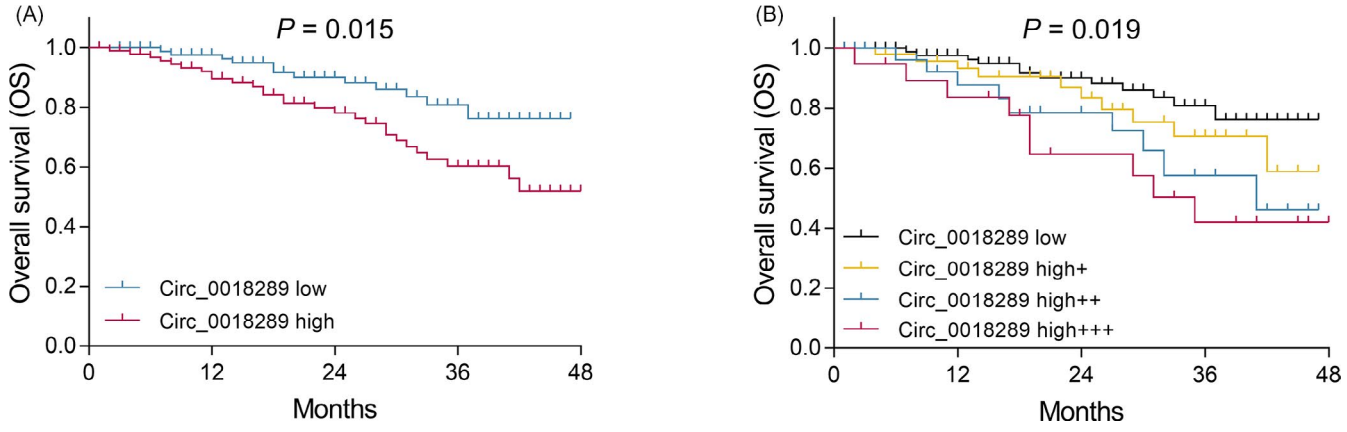

FIGURE 3 OS in patients with different tumor circ_0018289 expressions. The OS between patients with circ_0018289 high expression and patients with circ_0018289 low expression, and the OS among patients who had circ_0018289 low, high+, high++ as well as high+++ expression. circ, circular RNA; OS, overall survival

TABLE 3 Analysis of factors predicting DFS

	Cox's proportional hazard regression model			
			95% CI	
Items	*P* value	HR	Lower	Higher
Univariate Cox's regression				
Circ_0018289 high	.006	2.198	1.254	3.853
Age (≥45 y)	.168	1.462	0.852	2.508
HPV positive	.641	0.859	0.453	1.628
Histological type	.477	0.845	0.530	1.346
Higher pathological grade	<.001	2.655	1.771	3.980
Tumor size (≥4 cm)	.011	1.998	1.173	3.405
Lymph node metastasis	.005	2.231	1.281	3.884
Higher FIGO stage	<.001	3.315	1.903	5.775
Backward stepwise multivariate Cox's regression				
Circ_0018289 high	.042	1.816	1.022	3.226
Higher pathological grade	<.001	2.172	1.428	3.303
Higher FIGO stage	.023	2.008	1.100	3.666

Note: Factors predicting DFS were analyzed by univariate and backward stepwise multivariate Cox's proportional hazard regression model.

Abbreviations: CI, confidence interval; DFS, disease-free survival; FIGO, International Federation of Gynecology and Obstetrics; HPV, human papillomavirus; HR, hazard ratio.

TABLE 4 Analysis of factors predicting OS

	Cox's proportional hazard regression model			
			95%CI	
Items	*P* value	HR	Lower	Higher
Univariate Cox's regression				
Circ_0018289 high	.018	2.265	1.151	4.457
Age (≥45 y)	.222	1.493	0.785	2.842
HPV positive	.077	0.544	0.276	1.069
Histological type	.278	0.747	0.441	1.265
Higher pathological grade	<.001	3.477	2.028	5.963
Tumor size (≥4 cm)	.002	2.873	1.482	5.571
Lymph node metastasis	.008	2.386	1.257	4.529
Higher FIGO stage	<.001	4.106	2.050	8.226
Backward stepwise multivariate Cox's regression				
Higher pathological grade	.001	2.664	1.531	4.637
Tumor size (≥4 cm)	.013	2.337	1.197	4.562
Higher FIGO stage	.019	2.388	1.155	4.941

Note: Factors predicting OS were analyzed by univariate and backward stepwise multivariate Cox's proportional hazard regression model.

Abbreviations: CI, confidence interval; FIGO, International Federation of Gynecology and Obstetrics; HPV, human papillomavirus; HR, hazard ratio; OS, overall survival.

metastasis (*P* = .001), and FIGO stage (*P* = .001), while was not associated with age (*P* = .920), HPV status (*P* = .196), or histological type (*P* = .963) in cervical cancer patients (Supplementary Table S2).

4 | DISCUSSION

Cervical cancer is closely correlated with virus infection, and more than 90% of cervical cancer cases are developed from chronical infection of human papilloma virus (HPV). More importantly, multiple factors are involved in the development from HPV infection to cervical cancer.[11,12] Among all the factors, non-coding RNAs are increasingly reported, such as the microRNAs (miRNAs); however, despite that circRNAs are also illustrated as very promising factors in various carcinomas, relatively few studies of circRNAs in cervical cancer are reported.[13] As a consequence, we investigated the correlation of circ_0018289, a circRNA identified to be involved in pathogenesis of cervical cancer by our previous study, with clinical characteristics and prognosis in cervical cancer patients who underwent surgery, and found that: (a) circ_0018289 was upregulated in tumor tissue and could distinguish tumor tissue from adjacent non-tumor tissue; (b) circ_0018289 high expression in tumor tissue correlated with advanced clinical characteristics; and (c) circ_0018289 high expression in tumor tissue associated with unfavorable DFS/OS, and independently predicted worse DFS.

Several previous researches have paved the way for the investigation of circRNA functions in cervical cancer. For instance, a previous study illuminates that circ-MYBL2 enhances cancer cell proliferation and invasion by acting as a sponge of miR-361-3p in cervical cancer.[14] Furthermore, circ-AMOTL1 promotes cancer cell growth via elevating the expression of AMOTL1 of cervical cancer both in vivo and *in vitro*[15][11]. Another study reveals that circ_001038 promotes growth, migration, and invasion of cervical cancer cells through functioning as a competing endogenous RNA (ceRNA) of miR-337-3p.[16] Circ_0005576 inhibition represses growth, colony formation, and metastasis of cervical cancer cell lines HeLa cells and SiHa cells by sponging miR-153-3p that subsequently results in elevation of kinesin family member 20A (KIF20A) expression.[17] In addition, circ_0000745 acts as an oncogene in cervical cancer through promoting cancer cell proliferation, migration, and invasion.[18] To our best knowledge, the circ_0018289 was firstly reported in our previous study, which identified circ_0018289 as a dysregulated circRNA in cervical cancer tissues and cell lines by microarray, and

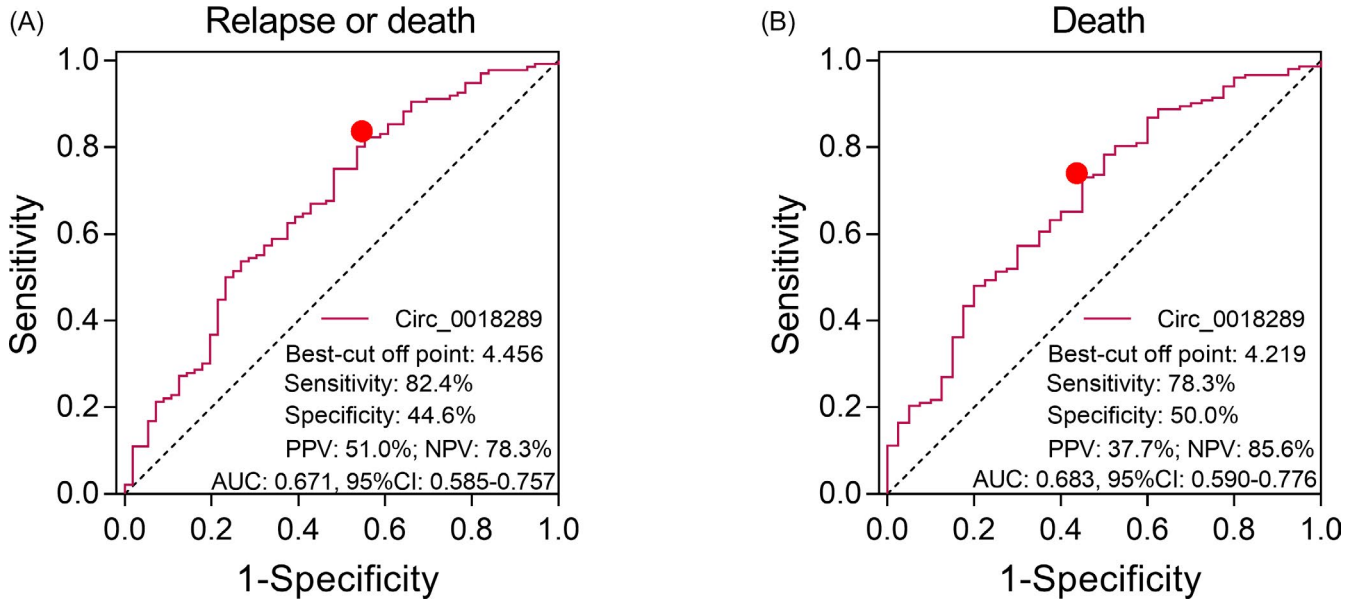

FIGURE 4 Predictive value of circ_0018289 expression in tumor tissue for DFS or OS. The ROC curve of the predictive value of circ_0018289 for relapse or death (A), the ROC curve of the predictive value of circ_0018289 for death (B). Circ, circular RNA; DFS, disease-free survival; OS, overall survival

further elucidated that circ_0018289 knockdown inhibited proliferation, migration, and invasion of cervical cancer cells via binding miR-497.[9] Furthermore, in our study, we found that circ_0018289 was overexpressed in tumor tissue and could clearly differentiate tumor tissue from non-tumor tissue, and tumor circ_0018289 high expression was correlated more worse clinical characteristics in cervical cancer patients. We hypothesized that these results might derive from the reason that circ_0018289 promoted cell proliferation, migration, and invasion of cervical cancer cells as discovered in our previous study, which could result in the progression of tumor, and subsequently contributed to the elevated circ_0018289 expression in tumor tissue and its positive correlation with more advanced clinical characteristics of cervical cancer patients.[9]

Besides the mechanistic role in etiology, here we also find several studies elucidating potential of circRNAs as biomarkers for cervical cancer management in the clinical practice. For instance, circSLC26A4 is found to be overexpressed in tumor tissue and cancer cells, and its high expression associates with unsatisfying survival profile in cervical cancer.[19] Another study reports that circ_0000745 high expression in tumor tissue correlates with poor differentiation in cervical cancer patients.[18] Circ-EIF4G2 is upregulated in tumor tissue and its elevated expression in tumor tissue associates with poor survival in cervical cancer patients.[20] Additionally, a study illuminates that circ_0001038 overexpression in tumor tissue is positively associated with lymph node invasion and myometrial invasion of patients with cervical cancer.[16] Moreover, there is a previous study elucidating that circ_0018289 is notably upregulated in cervical cancer tumor tissue compared with paired adjacent non-tumor tissue, which probably indicates that circ_0018 may have potential in serving as a biomarker for cervical cancer patients, which, however, needs to be validated by further studies.[9] In this study, we discovered that circ_0018289 high expression in tumor tissue was correlated with worse DFS and OS, and it was also an independent pejorative predictive factor for DFS. These results might be caused by that circ_0018289 could promote tumor progression by regulating cancer cell functions, which subsequently resulted in malignant tumor behaviors and caused disease progression that ultimately contributed to a worse survival in cervical cancer patients.[9] In addition, we also noticed that circ_0018289 independently predicted DFS but not OS, which might due to that circ_0018289 may participate more in the biological processes related to progression (main reason causing shorter DFS); in addition, the relatively small sample size may result in insufficient statistical power, which may also contribute to this result. Also, we would like to add a well-established circRNA that could serve as prognostic factor, whereas circRNA is still as relatively novel research area in oncology. Therefore, no well established circRNAs are found until now.

In addition, there were limitations that could not be ignored in this study. First, the sample size was relatively small, which might marginally reduce the statistical power. Second, the cervical cancer patients in our study were all resectable patients; thus, the value of circ_0018289 in unresectable patients, who were mostly in advanced FIGO stage, was not assessed. Third, the role of circulating expression of circ_0018289 in cervical cancer patients was not assessed, which, however, was due to that circ_0018289 expression in circulating sample is little. Last, the method for dividing circ_0018289 expression grade was not based on a reference and may be different from other studies researching circRNAs, which may result bias.

In conclusion, circ_0018289 could serve as a potential disease monitoring and prognostic biomarker in cervical cancer patients.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

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