PCAT-1 Expression is Associated With The Prognosis in Prostate Cancer.

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Research

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Abstract

Purpose: In the study, we aimed to estimate the prognostic significance of *PCAT-1* in patients with prostate cancer (PCa).

Methods: The expression of *PCAT-1* in paired PCa tissues and normal controls was examined via quantitative real-time polymerase chain reaction (qRT-PCR). The influence of *PCAT-1* level on clinical features was assessed using Chi-square test. The survival curves were plotted to estimate the prognosis of patients. And the Cox analysis was carried out to find promising predictive factors for patients.

Results: The expression level of *PCAT-1* in PCa tissues was significantly elevated compared with the adjacent normal control (P<0.0001). The increased expression of *PCAT-1* was affected by high Gleason score (P=0.017), positive serum PSA (P=0.011) and advanced TNM stage (P=0.003). The Kaplan-Meier survival curves showed that the overall survival rate of patients with high *PCAT-1* expression was significantly lower than those with low *PCAT-1* expression (P<0.001). Both univariate analysis (P=0.000, HR=10.623, 95%CI=5.798-19.464) and multivariate Cox regression analysis (P=0.000, HR=10.996, 95%CI=5.896-20.507) revealed that *PCAT-1* could act as a prognostic biomarker for PCa patients.

Conclusion: Taken together, overexpression of *PCAT-1* is involved in PCa progression and could be a potential prognostic biomarker for PCa patients.

Background

Prostate cancer (PCa) is one of the most common non-skin tumors among men, which is frequently detected in Europe and the United states with high malignancy, and especially occurs in men with the age of more than 50 years old [1, 2]. In addition, the incidence rate of PCa in developing countries and areas, such as China, has been reported to be increasing in recent years. PCa is one of the main reasons for a part of cancer-related deaths all around the world [3–5]. Chemotherapy, radiation and radical prostatectomy are currently the main treatments for PCa patients [6, 7]. Despite of the advantages and improvements on these curative treatments, the prognosis of PCa patients is still unsatisfactory. Moreover, prostate-specific antigen (PSA), TNM stage, and biopsy Gleason score are commonly taken as prognostic indicators at present [8, 9]. However, even with the same indicators the outcomes of patients are always diverse [10]. As a result, it is urgently required to find novel biomarkers that could exactly predict the outcomes of patients.

The human genome project study showed that in the transcription process,only a few genes (about 2%) are protein-coding, and the others are transcribed into non-coding RNAs (ncRNAs [11]. NcRNAs could be classified as short chain ncRNAs and long chain ncRNAs (lncRNAs) according to their size, and the latter account for the majority of ncRNAs [12, 13]. LncRNAs, which are previously regarded as “transcription noise”, are a class of non-coding RNAs with more than 200 nucleotides in length [14, 15]. It has been reported that lncRNAs are significantly involved in a variety of biological processes, such as chromatin modification, gene transcription, genetic imprinting, cell differentiation, apoptosis, protein activity

regulation, embryonic development and cancer development [16–18]. In addition, previous studies have demonstrated that approximately 15000 kinds of lncRNAs are found to be abnormally expressed in organism tissues. Prostate cancer-associated ncRNA transcripts 1 (*PCAT-1*) is a lncRNA that was originally identified in prostate cancer.

In the present study, we aimed to compare the expression levels of lncRNA *PCAT-1* in PCa tissues and paired normal controls and to identify its role in the prognosis of PCa patients.

Materials And Methods

Patients and specimens

Our retrospective study recruited 129 patients as objects, who were pathologically diagnosed with PCa in Zhongnan Hospital of Wuhan University, age range of 25 to 75 years. The tissue samples were immediately prepared and stored in liquid nitrogen after surgical resection. Patients received no chemo- or radio- therapy before surgery. The information of patients is updated every 3 months for a 5-year follow-up. The study was carried out with the admission of Ethics Committee in Zhongnan Hospital of Wuhan University. And the informed consent was provided by each participant.

Quantitative real-time polymerase chain reaction (qRT-PCR)

Total RNA was isolated from tissue samples using the Trizol Reagent (Invitrogen) according to the manufacture's instructions. The RNA was reverse transcribed into the first strand of cDNA with a Reverse Transcription Kit (Takara). Then quantitative real-time PCR was conducted using the ABI PRISM 7000 Fluorescent Quantitative PCR system (Applied Biosystems, Foster City, USA). The expression of *PCAT-1* was determined using the $2^{-\Delta\Delta Ct}$ method and normalized to the internal control *GAPDH*.

Statistical analysis

All data were analyzed by Sigmaplot 12.5 (Systat Software Inc, CA, USA) and SPSS 18.0 (International Business Machines Corporation, USA) software. The t-test was conducted to compare the expression difference of *PCAT-1* in PCa tissues and paired normal controls. The χ^2 test was used to describe the associations between *PCAT-1* levels and patients' clinical characteristics. Kaplan-Meier method was adopted to evaluate the overall survival rate of patients. The prognostic biomarkers for PCa patients were identified using Cox regression analysis. *P* values less than 0.05 were considered as statistical significant.

Results

Up-regulation of *PCAT-1* in PCa tissues

The expression of *PCAT-1* in PCa tissues and corresponding normal controls was determined using qRT-PCR. As shown in Fig. 1, the level of *PCAT-1* in PCa tissues was 2.91 ± 0.70 (mean ± SD), while that in normal controls was 1.72 ± 0.57. The *PCAT-1* expression was significantly increased in PCa tissues compared with normal controls ($P < 0.0001$).

The association between *PCAT-1* expression and clinical parameters of PCa patients

To explore the effects of clinical parameters on *PCAT-1* expression, the Chi-square test was performed. Patient were divided into two groups based on the median value of *PCAT-1* levels: the high expression group (n = 65) and the low expression group (n = 64). According to the results of χ^2 test, *PCAT-1* up-regulation was associated with high Gleason score ($P = 0.017$), positive serum PSA ($P = 0.011$) and advanced TNM stage ($P = 0.003$) (Table 1). On the other hand, *PCAT-1* expression shared no close relationship with age ($P = 0.250$), NED rate ($P = 0.093$), urine retention ($P = 0.179$) and hematuria ($P = 0.135$).

Table 1
Relationship between *PCAT-1* expression and clinical characteristics of PCa patients

Clinical features	Case NO.	Expression		χ^2	*P* value
		High	Low		
Age				1.325	0.250
≦ 55	68	31	37		
> 55	61	34	27		
NED rate				2.814	0.093
≦ 40%	67	29	38		
> 40%	62	36	26		
Urine retention				1.806	0.179
Present	56	32	24		
Absent	73	33	40		
Hematuria				2.233	0.135
Ever	69	39	30		
Never	60	26	34		
Gleason score				5.680	0.017
≦ 7	57	22	35		
> 7	72	43	29		
Serum PSA				6.517	0.011
Positive	65	40	25		
Negative	64	25	39		
TNM stage				8.548	0.003
I,II	70	27	43		
III,IV	59	38	21		

The correlation between *PCAT-1* expression and prognosis of patients

The Kaplan-Meier curves and Cox regression analysis were conducted to evaluate the prognostic value of *PCAT-1* in PCa. During the 5-year follow-up, 56 of 65 (86.15%) patients with high *PCAT-1* expression died, while only 14 (21.88%) cases with low *PCAT-1* expression died. As shown in Fig. 2, the overall survival rate of patients with high *PCAT-1* expression was significantly lower than those with low *PCAT-1* expression ($P < 0.001$). Furthermore, univariate Cox regression analysis revealed that urine retention ($P = 0.015$, HR = 1.790, 95%CI = 1.119–2.866), Gleason score ($P = 0.000$), serum PSA ($P = 0.036$), TNM stage ($P = 0.026$) and *PCAT-1* expression ($P = 0.000$) were related with the prognosis of PCa patients (Table 2). Meanwhile, the multivariate Cox regression analysis further demonstrated that urine retention ($P = 0.008$, HR = 1.922, 95%CI = 1.186–3.113), Gleason score ($P = 0.001$, HR = 2.507, 95%CI = 1.469–4.278) and *PCAT-1* expression ($P = 0.000$, HR = 10.996, 95%CI = 5.896–20.507) were prognostic biomarkers for PCa patients.

Table 2
Univariate and multivariate analysis of risk factors

Clinical features	Univariate		Multivariate	
	P value	HR(95%CI)	*P* value	HR(95%CI)
Urine retention	0.015	1.790(1.119–2.866)	0.008	1.92(1.186–3.113)
Gleason score	0.000	2.634(1.565–4.433)	0.001	2.507(1.469–4.278)
Serum PSA	0.036	1.665(1.034–2.681)	-	-
TNM stage	0.026	1.705(1.066–2.729)	-	-
PCAT-1 expression	0.000	10.323(5.798–19.464)	0.000	10.996(5.896–20.507)

Discussion

PCa is one of the most important malignancies in the male reproductive system, and also a common malignant tumor in men. Its biological behavior is complicated and its pathological process is difficult to grasp. An increasing number of investigations have demonstrated that the occurrence and development of PCa is controlled and regulated by multiple indicators, such as daily habit, environmental factors and gene variation [19]. In recent years, the incidence rate of PCa has increased remarkably in China. To improve the treatment efficacy and the outcomes of patients, numerous studies are carried out to seek relative prognostic biomarkers for PCa. Mavridis et al. showed that *miR-224* down-regulation was related with poor prognosis of PCa patients [20]. In the study of Zheng et al., they reported that *SFRP1* could act as a prognostic marker for PCa patients [21]. However, the etiology and tumorigenesis of PCa has not been fully elucidated yet. Therefore, searching novel biomarkers is requested to understand the PCa pathogenesis and improve the patients' outcomes.

LncRNAs have increasingly been linked to many human diseases. The diversity and nucleotide length of lncRNAs allow them to possess a broad range of biological functions despite the lack of protein-coding

potential. *PCAT-1*, a newly confirmed lncRNA, is mapped to human chromesome 8q24 desert and is significantly contributed to the development and progression of cancers [22, 23]. It is reported by Zhao et al. that lncRNA *PCAT-1* up-regulation could enhance the cell proliferation and migration in non-small cell lung cancer [24]. Prensner et al. found that *PCAT-1* expression could generate a functional deficiency of homologous recombination in cancers through targeting *BRCA2* [25]. The study carried out by Yan et al. demonstrated that the expression of lncRNA *PCAT-1* was significantly elevated in hepatocellular carcinoma tissues compared with adjacent normal controls and this over-expression was related with poor prognosis of patients [26]. Furthermore, Prensner et al. claimed that lncRNA *PCAT-1* could stimulate cell proliferation in PCa through regulation cMyc [16]. However, the clinical role fo lncRNA *PCAT-1* in PCa patients was still unclear.

In the present study, the correlation between *PCAT-1* expression and prognosis of PCa patients was evaluated. First, we determined and compared the expression of lncRNA *PCAT-1* in PCa tissues and the corresponding normal controls using qRT-PCR and t-test. The results showed a significantly high expression of *PCAT-1* in PCa tissues compared to adjacent normal controls, which was consistent with the previous studies. Chi-square test was carried out to explore the potential biomarkers influencing *PCAT-1* expression. Gleason score, serum PSA and TNM stage were finally proved to play important roles in the expression of *PCAT-1*. Based on these findings, we concluded that *PCAT-1* is closely related with the development and progression of PCa, and thus we hypothesized that *PCAT-1* might be involved in the prognosis of PCa patients. The Kaplan-Meier curves revealed that patients with low *PCAT-1* expression had a higher survival rate than those with high expression. Furthermore, the Cox regression analysis showed that *PCAT-1* was a predictive candidate biomarker for the prognosis of PCa patients.

Clinically, our study confirmed the over-expression of lncRNA *PCAT-1* in PCa tissues compared to normal controls. lncRNA *PCTA-1* overexpression has been implicated in the poor prognosis of PCa. While the direct therapeutic targeting of lncRNAs has not been proven and how *PCAT-1* affects the development and progression of PCa is still needed more investigations with larger sample size.

Conclusions

In conclusion, our study confirmed the over-expression of lncRNA *PCAT-1* affected the cancer development and progression in PCa and lncRNA *PCAT-1* might be a valuable prognosis biomarker for patients. Moreover, our study identified the prognostic role of lncRNA *PCAT-1* for PCa patients for the first time.

List Of Abbreviations

prostate cancer (PCa)

quantitative real-time polymerase chain reaction (qRT-PCR)

non-coding RNAs (ncRNAs)

long chain ncRNAs (lncRNAs)

Prostate cancer-associated ncRNA transcripts 1 (*PCAT-1*)

Transcription Kit (Takara)

Declarations

Ethics approval and consent to participate

This study was supported by the Ethics Committee of Zhongnan Hospital of Wuhan University and also has been carried out in accordance with the World Medical Association Declaration of Helsinki.

The subjects had been informed the objective. Certainly, written consents were signed by every subject in this study.

Consent for publication

We obtaining permission from participants to publish their data.

Availability of data and materials

Data sharing is not applicable to this article as no datasets were generated or analysed during the current study.

Conflict of Interest

The authors declare that they have no conflict of interest.

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The authors did not receive support from any organization for the submitted work.

Authors' contributions

S.L. design of the work; S.L. the acquisition, analysis, X.R. interpretation of data; X.R. the creation of new software used in the work; T.L. have drafted the work or substantively revised it. All authors read and approved the final manuscript.

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Figures

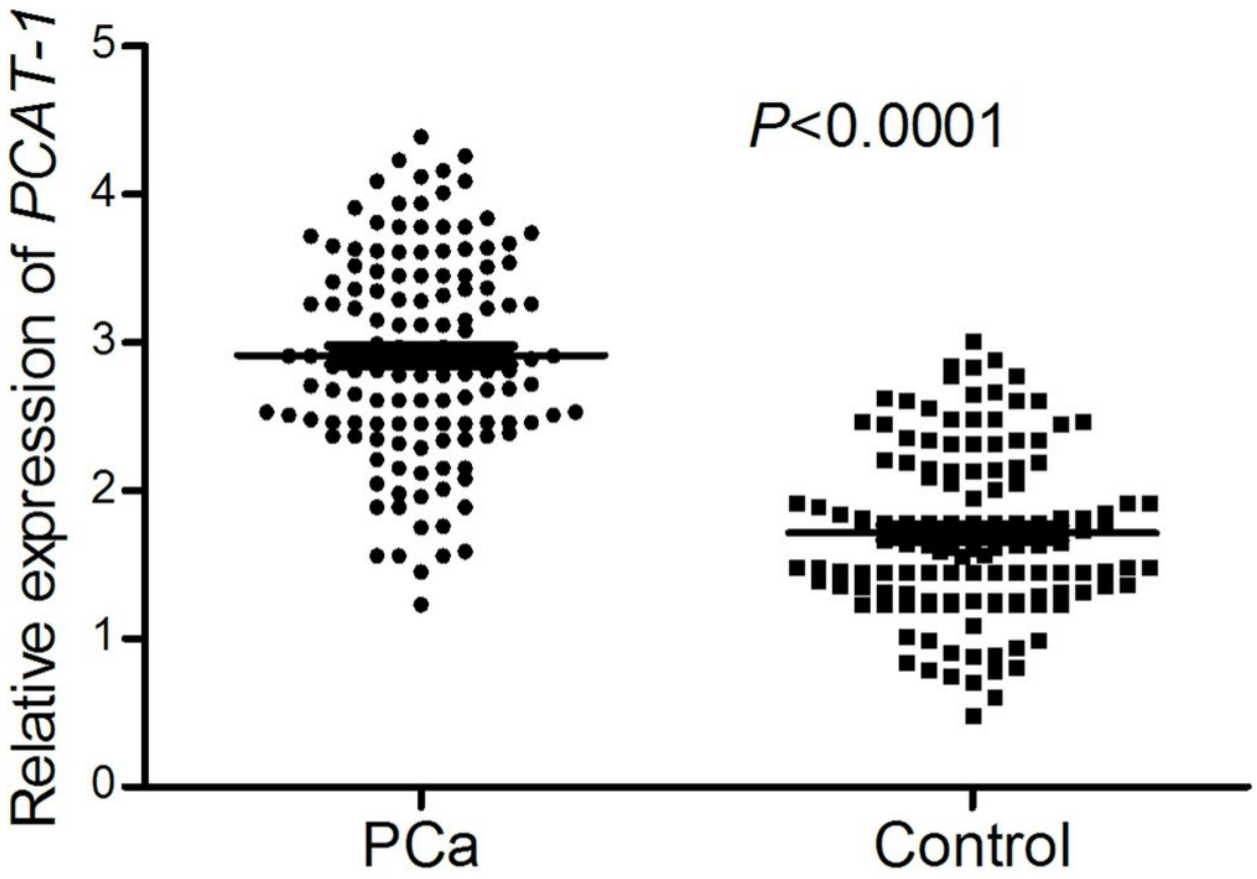

Figure 1

The expression of PCAT-1 in PCa tissues and adjacent normal controls. The PCAT-1 level was significantly higher in PCs tissues than the paired normal controls (P<0.0001).

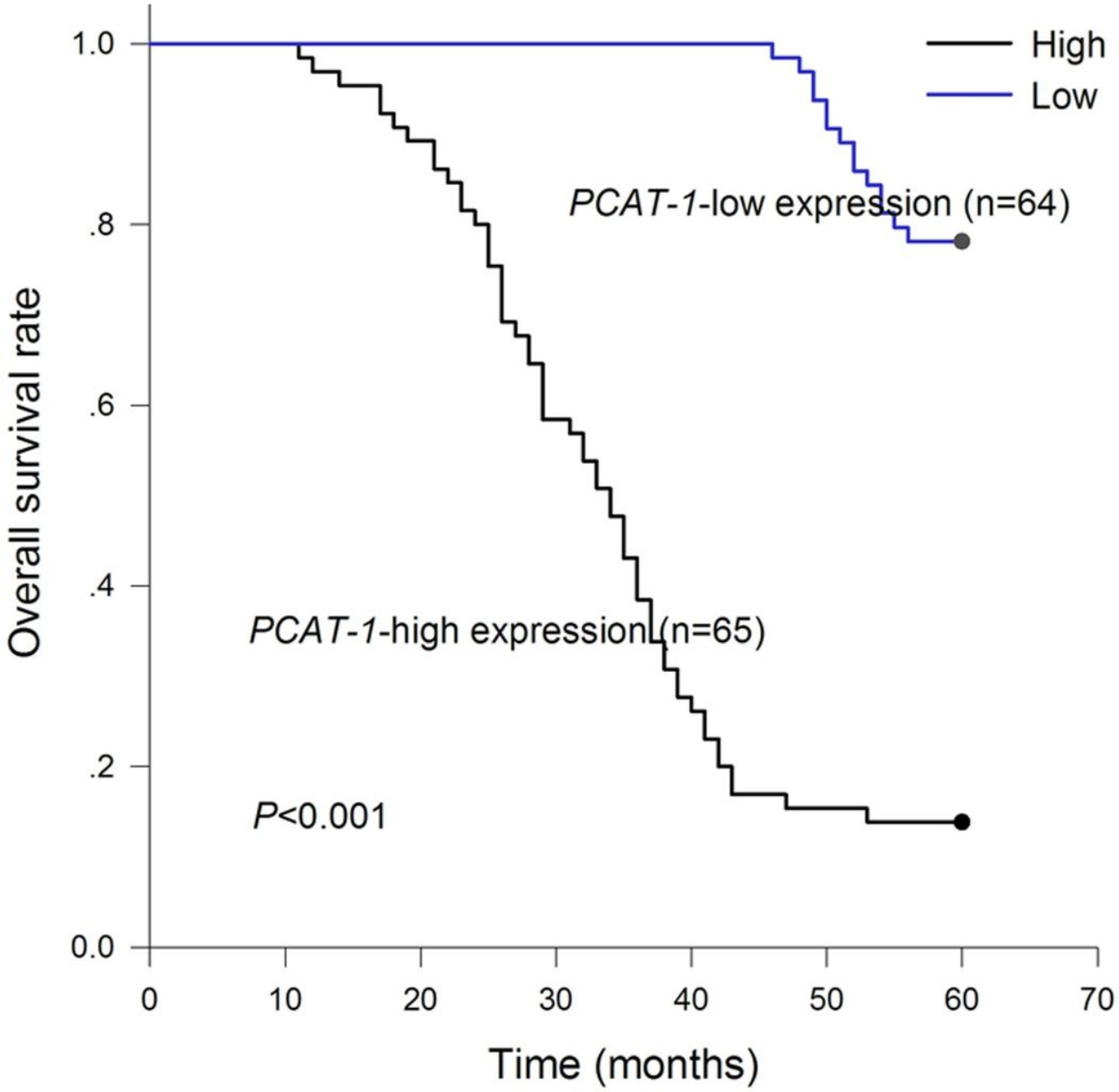

Figure 2

Kaplan-Meier curves for PCa patients with diverse PCAT-1 expression. It was concluded that patients with low PCAT-1 expression lived longer than those with high PCAT-1 expression (P<0.001).