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LncRNA RP11-597D13.9 expression and clinical significance in serous Ovarian Cancer based on TCGA database

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Abstract: **To analyze the relation between the expression of RP11-597D13.9 and prognosis in serous ovarian cancer (SOC) patients via The Cancer Genome Atlas (TCGA) database, we downloaded clinical information of SOC patients from TCGA and downloaded the lncRNA expression data from The Atlas of Noncoding RNAs in Cancer (TARNIC) database. 2 and t test were used to analyze the association of RP11-597D13.9 and the clinicopathological features of SOC patients. Kaplan-Meier was used to analyze the relationship between the expression of RP11-597D13.9 and the overall survival of SOC patients. Univariate and multivariate COX regression was further used to analyze whether RP11-597D13.9 was an independent prognosis marker of SOC patients. Our results showed that RP11-597D13.9 expression level was not associated with age, TNM and grade but associated with survival status (P=0.002) and race (p=0.049). Compared with patients with low expression level, SOC patients with high RP11-597D13.9expression level had shorter overall survival time. Besides this, the RP11-597D13.9 expression level in deceased patients significantly higher than living patients. Univariate and multivariate COX regression analysis showed that age and RP11-597D13.9 are the indicators of the prognosis of patients with SOC. Therefore, positive expression of RP11-597D13.9 could be an important independent marker of poor prognosis in patients with SOC.**

Keywords: R11-597D13.9, Serous Ovarian Cancer, prognosis, The Cancer Genome Atlas

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1. Introduction

Ovarian cancer is the fourth most common gynecologic malignancy in women and is currently the most deadly tumor. Due to the lack of effective early diagnostic methods, 70% of the patients were diagnosed with advanced tumors[1]. Epithelial ovarian cancer (EOC) account for 90% of all ovarian cancer cases and is generally diagnosed at advanced stage[2]. Despite the advances in surgical techniques and conventional chemotherapy, the prognosis of EOC patients has not obviously improved. The long-term survival (5 years or more) is still less than 30%[3].

With the development of whole-genome sequencing technology, it was determined that more than 98% of the mammalian genome was noncoding RNAs[4]. Among them are long noncoding RNAs (lncRNA), which are more than 200nt in length and unable to be translated into proteins[5]. LncRNA is regarded as a new regulator in numerous biological process and has attracted more and more researchers' attention. A large number of lncRNAs exhibit a tissue- specific pattern and display weaker evolutionary constraints and lower expression levels than protein-coding genes[6,7]. Therefore lncRNA was considered as "transcription of garbage" before and caused less attention. However, more and more studies have indicated that lncRNAs play an important role during the development of tumor, including EOC. For example, HOTAIR[8], AB073614[9] and ANRIL[10] has been proved they are served as pivotal regulators in the biological process of EOC.

In order to further search the important marker of poor prognosis of EOC patients, we analyzed the lncRNA expression and clinical information of serious ovarian cancer (SOC) patients, one of the most common subtype of EOC, through the database of TCGA[11] and TARNIC[12]. In this study, we first analyzed the relationship between RP11-597D13.9 and the clinicopathological features of SOC patients. The relationship between the expression level of lncRNA RP11-597D13.9 and the prognosis of SOC patients was next analyzed by *Kaplan-Meier* method, and we also further clarify the expression of RP11-597D13.9 and the prognosis of SOC patients by univariate and multivariate Cox regression method.

2. Materials and Methods

2.1. Retrieval and integration of public data

Clinical information about 604 SOC patients was downloaded from the TCGA database (http: //cancergenome.nih.gov). The lncRNA expression levels of SOC patients were downloaded from TANRIC database (http://ibl.mdanderson.

org/tanric/_design/basic/index.html).

The VLOOKUP function in excel was used to combine the clinical data and the lncRNA RP11-597D13.9 expression according to the ID number. 192 SOC patients were excluded because of lack of RP11-597D13.9 expression data and 7 patients also excluded due to incomplete primary clinical data. Finally, 405 SOC patients with the data of RP11-597D13.9 expression were included.

2.2. Statistical analysis for survival and correlation

For quantitative data, all results are expressed as the Mean ± standard deviation (SD) and statistical significance between groups was determined using a student's t test. For count data, all results are expressed as rate (%) and statistical significance between groups were determined using a Chi-square test (χ^2). Due to the expression data of RP11-597D13.9 with 405 SOC patients failed to meet normal distribution. Median value was used as demarcation point to divide patients into high-expression and low expression groups. Overall survival analysis was calculated by the Kaplan-Meier method, and the log-rank test was utilized to compare the differences between patient groups. Univariate and multivariate Cox proportional hazards regression analysis was further carried out. The statistical analysis of this study was performed using SPSS 20.0 (SPSS, Inc., Chicago, IL, USA).

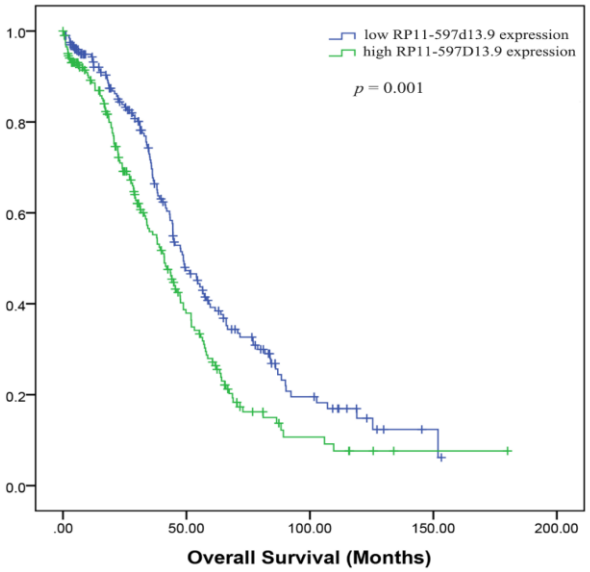

Figure 1. The relation between RP11-597D13.9 expression and overall survival of SOC patients.

Table 1. The correlation of RP11-597D13.9 expression with clinicpathological features

clinicpathological features		No. of patients	RP11-597D13.9 expression		P
			Low group	High group	
Age		405			0.215
	≤50		55(55.6%)	44(44.4%)	
	>50		146(47.7%)	160(52.3%)	
TNM stage		404			0.179
	II		7(31.8%)	15(68.2%)	
	III		162(50.5%)	159(49.5%)	
	IV		31(50.8%)	30(49.2%)	
Grade		397			0.849
	G1/G2		23(47.9%)	25(52.1%)	
	G3		175(50.1%)	174(49.9%)	
Site		381			0.251
	Left		26(50.0%)	26(50.0%)	
	Right		28(60.9%)	18(39.1%)	
	Bilateral		135(47.7%)	148(52.3%)	
Invasion		156			0.282
	YES		47(45.2%)	57(54.8%)	
	NO		29(55.8%)	23(44.2%)	
OS status		405			**0.002***
	LIVING		45.2 ±40.6	34.9 ±36.9	
	DECEASED		43.8 ±27.8	34.4 ±23.3	
Race		393			**0.049***
	White		171(47.9%)	186(52.1%)	
	Others		24(66.7%%)	12(33.3%)	

*P<0.05 indicates the significant difference.

3. Results

3.1. The correlation of RP11-597D13.9 expression with clinicpathological features

Due to the skewed distribution of the RP11-597D13.9 with 405 SOC patients, we used the median value (0.0476) as the demarcation. Patients with RP11-597D13.9 expression below the median were classified into the low expression group and the patients with RP11-597D13.9 expression higher than the median were classified into the high expression group. The association between the RP11-597D13.9 expression and clinicopathological features was performed. As shown in table 1, the RP11-597D13.9 expression was not associated with age, TNM stage, grade, site and invasion. However, the RP11-597D13.9 expression was correlated with survival status (P =0.002<0.05) and race (P =0.049 <0.05).

3.2. The relationship between RP11-597D13.9 expression and overall survival of SOC patients

Since the expression of RP11-597D13.9 is mainly related to survival status, we firstly analyzed the relationship between of RP11-597D13.9 expression and overall survival of SOC patients based on TCGA database. Kaplan–Meier survival analysis indicated that the high expression group had poor survival as compared to the low expression group (P=0.002 <0.05) (Figure 1).

Table 2. Univariate and multivariate Cox regression analyses of overall survival

Factors	Univariate Cox			Multivariate Cox		
	HR	95%CI	P	HR	95%CI	P
Age	1.022	1.010-1.034	**<0.001***	1.025	1.013-1.038	**<0.001***
Grade	1.148	0.79-1.667	0.469	1.182	0.79-1.769	0.417
TNM	1.262	0.959-1.659	0.097	1.259	0.942-1.684	0.120
Race	1.342	0.856-2.106	0.200	1.676	1.051-1.672	**0.030***
RP11-597D13.9	1.472	1.148-1.888	**0.002***	10.471	1.313-83.491	**0.027***

*P<0.05 indicates the significant difference.

In addition, we further analyzed the expression levels of RP11-597D13.9 between living SOC patients and the deceased SOC patients. As shown in Figure 2, the RP11-597D13.9 expression level in tumor tissues of deceased SOC patients significantly higher than living SOC patients. These results indicated that high expression of RP11-597D13.9 predicts shorter survival time of SOC patients.

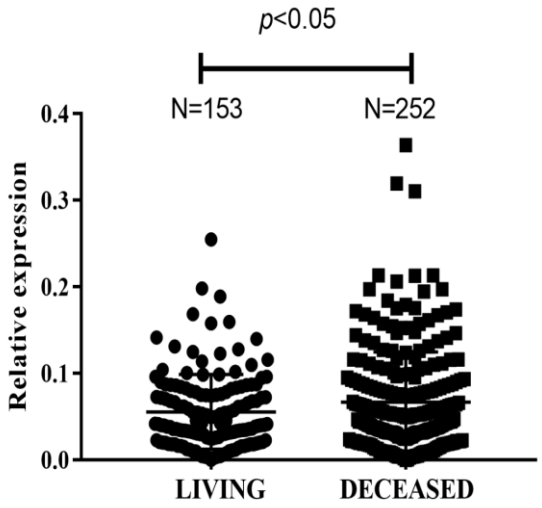

Figure 2. Differential expression of RP11-597D13.9 between living and deceased patients of SOC.

Univariate and multivariate Cox analysis also was performed to check the independent marker of SOC patients' overall survival. In univariate Cox analysis, age (HR=1.022, 95%CI, 1.010 to 1.034, P<0.001) and the expression of RP11-597D13.9 (HR=1.472, 95%CI, 1.148 to 1.888, P <0.001) maybe the prognostic marker of SOC. In multivariate analysis, all variables considered as potential prognostic factors were included in a Cox proportional hazards regression models to identify independent prognostic factors. The results showed that for overall survival analysis of SOC patients, age (HR=1.025, 95%CI, 1.013 to 1.038, P <0.001) and RP11-597D13.9 (HR=10.471, 95%CI, 1.313 to 83.491, P =0.027) signature were independent predictor (Table 2).

4. Discussion

In recent years, lncRNAs has been as the hot topic of research fields. As the tissue specificity, lncRNA is increasing concerned by various fields, especially in the cancer fields[6,13,14]. Previous studies reported that lncRNA could serve as the prognosis marker for cancer patients, including EOC patients. For example, high expression of LINC00092[15], HOTAIR[8] and ASAP1-IT1[16] predicts poor prognostic in patients with ovarian cancer. In our study, we analyzed the correlation of RP11-597D13.9 expression and the overall survival of SOC patients based on TCGA

database. Our results indicated that RP11-597D13.9 expression was an independent marker of SOC patients for overall survival. To the best of our knowledge, this is the first report on the correlation of RP11-597D13.9 expression and the overall survival of SOC patients. This study may provide some guidance for the prognosis of SOC patients.

More than 3000 lncRNAs have been identified so far. However, only 1% of lncRNAs have been shown to have important roles in various biological processes[17]. Given lncRNAs are poorly conservative and highly versatile in modulating biomolecules, which may be more sensitive to disease change. Therefore, there is an urgent need to identify new and important lncRNA in cancer. TCGA database is the best research tool for studying the relationship between lncRNA and cancer with RNA-seq profiles and clinicopathological features of cancer patients. The TCGA project is a large-scale study using the high-throughput sequencing technology to detect genomes expression of various cancers[11]. Up to now, there are more than 30 kinds of cancers have covered about 10,000 cases samples in TCGA project. The advantage of this study lies in the use of a large sample of relatively complete SOC patient information in the TCGA database to analyze the relationship between the expression level of RP11-597D13.9 and the overall survival of SOC patients, which is expected to be an important marker of poor prognosis of SOC.

However, although we did our best to analyze, there are still some limitations in our study. Firstly, most patients that included in TCGA were white people, Asians and others people are less. Due to the different expression of lncRNA among different races, this study may be more suitable for white patients with SOC. In table 2, Multivariate Cox regression analysis showed that RP11-597D13.9 expression was correlated with race maybe due to less Asian and others patients. This also maybe the reason of difference result with our previous study[18]. Secondly, not all patients have complete clinical data. In table 1, some SOC patients lack the information of lymphatic metastasis and tumor sites, resulting in inconsistent conclusions. Meanwhile, in table 2, Cox analysis was performed to check whether the age, TNM stage, grade, race and RP11-597D13.9 expression was the independent marker of SOC patients' overall survival. Age, TNM stage and grade had been demonstrated as the independent marker of cancer[19-21]. However, in our study, age was the independent marker of SOC patients' overall survival and TNM stage and grade was not. The reason may some patients lack the TNM stage and grade features. Therefore, further studies are needed to assess the relationship between RP11-597D13.9 and ovarian cancer.

5. Conclusion

RP11-597D13.9 expression was an independent marker of SOC patients for overall survival. To the best of our knowledge, this is the first report on the correlation of RP11-597D13.9 expression and the overall survival of SOC patients.

Acknowledgments

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