LncRNA PCAT-1 in gastrointestinal cancers
A meta-analysis
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Abstract
Background: Prostate-cancer-associated ncRNA transcript 1 (PCAT-1), a newly discovered lncRNA, was implicated in the progression of multiple tumors. We conducted a systematic review and meta-analysis to determine its prognostic potential for gastrointestinal cancers.

Methods: A literature survey was conducted by searching the PubMed, Web of Science, Cochrane Library, Embase together with Wanfang, and China National Knowledge Infrastructure database for articles published as of October 15, 2017. Hazard ratio (HR) or odds ratio (OR) with 95% confidence intervals (95% CIs) were calculated to demonstrate prognostic value of PCAT-1 using Stata 12.0 software.

Results: A total of 6 studies with 961 cases were pooled in the analysis to evaluate the prognostic value of PCAT-1 in gastrointestinal cancers. Increased PCAT-1 expression was significantly correlated with poor overall survival (OS) (HR = 1.04, 95% CI: 1.02–1.06). Statistical significance was also observed in subgroup meta-analysis stratified by cancer type, histology type, sample size, and analysis type. Additionally, high expression of PCAT-1 was significantly associated with deeper tumor invasion (OR = 4.46, 95% CI: 3.00–6.63), positive lymph node metastasis (OR = 3.76, 95% CI: 1.39–10.16), and advanced clinical stage (OR = 4.09, 95% CI: 1.55–10.82).

Conclusion: High expression of PCAT-1 was related to poor prognosis and could be a promising biomarker of clinicopathologic features in gastrointestinal cancers. More studies will be necessary to verify and strengthen the clinical value of PCAT-1 in gastrointestinal cancers.

Abbreviations: 95% CI = 95% confidence interval, CNKI = China National Knowledge Infrastructure, CRC = colorectal cancer, ESCC= esophageal squamous cell carcinoma, GC = gastric cancer, HCC = hepatocellular carcinoma, HE = high expression, HR = hazard ratio, MVA = multivariate analysis, NA = not available, NOS = Newcastle–Ottawa scale, OR = odds ratio, OS = overall survival, PCAT-1 = prostate-cancer-associated ncRNA transcripts 1, RT-qPCR = real-time quantitative polymerase chain reaction.

Keywords: gastrointestinal cancer, lncRNA, meta-analysis, prostate-cancer-associated ncRNA transcript 1, prognosis

1. Introduction
Gastrointestinal malignancies are the major and complex diseases in the world, which have caused a great burden on human health, families, and society.[1,2] They originated from the gastrointestinal tract or accessory organs of digestion. The patients diagnosed with such kind cancers usually have an unfavorable prognosis, especially for 5-year survival rate. Over the past few decades, many studies have focused on searching promising novel biomarkers for gastrointestinal cancers.[3-5] Exploring early diagnosis and prognosis tumor-markers is important and also in urgent need.

Long noncoding RNAs (lncRNAs), as rising stars in recent years, have attracted mountains of attention for their vital roles in diverse biologic processes.[6-8] Through lncRNAs are a group of noncoding RNAs with over 200 nucleotides in length but without protein-coding ability, more and more lncRNAs were identified and reported to function as oncogene or tumor suppressor factor in tumorigenesis and cancer progression.[9-11] Furthermore, they might act as diagnostic, prognostic biomarkers, or therapeutic targets in human cancers.[12-15]

Prostate-cancer-associated ncRNA transcript 1 (PCAT-1) was a novel identified IncRNA. It was firstly found in prostate cancer and was reported playing an active role in promoting prostate cancer cell proliferation.[16] In recent years, abnormal expression of PCAT-1 was found in multiple cancers and involved in the progression of various tumors, such as breast cancer, bladder cancer, glioblastoma, and nonsmall-cell lung cancer.[17-20] Notably, the role of PCAT-1 in gastrointestinal cancers has aroused considerable interest. Researchers reported that PCAT-1 was implicated in tumor invasion and metastasis,[21-23] and found correlations between PCAT-1 expression and clinical outcomes in multiple gastrointestinal malignancies.[24-26] However, until now there was no meta-analysis systematically elucidating the prognostic value of PCAT-1 in gastrointestinal tumors, and considering the limitations associated with specimen
sizes or study methodology of the single study. Consequently, we performed this study to explore the clinical values of PCAT-1 in gastrointestinal cancers by gathering all related published data.

2. Materials and methods

2.1. Publication search

Since this is a meta-analysis, ethical approval was not needed. To identify potentially eligible articles, a comprehensive literature search of PubMed, Web of Science, Cochrane Library, Embase together with Wanfang, and China National Knowledge Infrastructure (CNKI) database was performed prior to October 15, 2017. The following keywords were used for searching: PCAT-1 or PCA1 or PCAT1 or prostate-cancer-associated ncRNA transcript 1 or prostate-cancer-associated transcript-1. The reference lists of relevant literature were manually searched for additional eligible articles.

2.2. Inclusion and exclusion criteria

Inclusion criteria are as the following: studies detecting the expression of PCAT-1 in gastrointestinal cancers; the association between PCAT-1 expression and overall survival (OS) was investigated; sufficient survival data were provided for the hazard ratio (HR) with 95% confidence interval (CI); and patients were divided into 2 groups.

The following studies were excluded: duplicate publications; those on non gastrointestinal tumors, or animal experiments; studies investigating the molecular structure and functions of PCAT-1 without survival outcome; and reviews, letters, case reports, conference abstracts, or editorials.

2.3. Data extraction

The following data and information were collected from all eligible studies: publication information: name of first author, publication year, country; Patients’ characteristics: cancer type, number of patients, expression pattern, follow-up duration, gender, histologic grade, tumor depth, lymph node metastasis, distant metastasis and clinical stage; PCAT-1 expression measurement and cut-off value; and HRs of PCAT-1 for OS as well as their 95% CIs and P-values.

If only Kaplan–Meier curves were available, we extracted data from the graphical survival plots and estimated the HRs. If a study reported the data in multivariate analysis or/and univariate analysis for OS, the former was directly applied.

The Newcastle–Ottawa quality assessment scale (NOS) used to assess the quality of enrolled studies, with the score ranging from 0 to 9 points in the method, A study with a score ≥6 was considered high quality.

Figure 1. Flow diagram of the study search and selection process.
2.4. Statistical analysis

All statistical analyses were executed using STATA statistical software version 12.0 (STATA, College Station, TX) in this meta-analysis. Heterogeneity across studies was quantified with the $I^2$ statistics and Cochran $Q$ test. The random-effects model was conducted to analyze the relationship between PCAT-1 expression and clinical outcomes when calculated $I^2$ values $>50\%$ or $P < .1$. If there was no significant heterogeneity among studies, the fixed effects model was applied. Probable publication bias was displayed by constructing a funnel plot and conducting Begg test. Sensitivity analysis was used to evaluate the robustness of the pooled results. A $P$-value of $<.05$ was considered statistically significant.

3. Results

3.1. Included literatures

As shown in the flow diagram (Fig. 1), a total of 135 studies were initially identified as appropriate from PubMed, Web of Science, Cochrane Library, Embase, Wanfang, and CNKI database. After excluding duplicates, 36 records were reserved. And after carefully screening those titles and abstracts, 23 irrelevant articles were removed. From the 13 remaining articles, 7 were excluded because of incomplete data or absence of survival outcome. Ultimately, a total of 6 studies$^{[24-29]}$ were included in this meta-analysis according to the selection criteria.

3.2. Characteristics of the enrolled studies

The main features of the 6 enrolled studies are summarized in Table 1. All those publications were written in English with the released period from 2013 to 2017. There were totally 961 patients with median sample size of 160.2, with a wide range from 108 to 321. Four different kinds of gastrointestinal cancers were evaluated in this meta-analysis: 2 esophageal squamous cell carcinoma (ESCC), 2 gastric cancer (GC), 1 colorectal cancers (CRCs), and 1 hepatocellular carcinoma (HCC). All detected samples were fresh or frozen tissues from the patients without any preoperative treatments. The expression of PCAT-1 was measured by RT-qPCR. All are retrospective studies regarding relevance between PCAT-1 expression and gastrointestinal cancers prognosis. In this meta-analysis, the quality scores of all eligible studies were varied from 6 to 9, with a mean value of 7.5.

3.3. Results of the meta-analysis

3.3.1. Relationship between lncRNA PCAT-1 and OS.

All included studies comprising 961 patients reported the relationship between lncRNA PCAT-1 and OS in gastrointestinal cancers. No significant heterogeneity across-studies was found ($I^2=40.6\%$; $P_{I^2}=.135$), so the fixed effects model was used to estimate the pooled HR. The pooled results showed that high expression of PCAT-1 in cancer tissues was significantly correlated with poor OS in gastrointestinal cancers (HR $= 1.04$, 95% CI: 1.02–1.06, $P < .001$) (Fig. 2). The patients with high PCAT-1 had a worse OS than those with low expression of PCAT-1. PCAT-1 might be a significant prognostic factor of OS for gastrointestinal cancer patients.

3.3.2. Subgroup analysis of PCAT-1 in OS.

Subgroup analyses for OS were also performed. As the results showed in Table 2, compared with the merged HR, high PCAT-1 showed a stronger association with unfavorable OS in the subgroups of GC (HR $= 1.05$, 95% CI: 1.02–1.08, $P < .001$), and ESCC (HR $= 1.04$, 95% CI: 1.01–1.06, $P < .001$). In addition, the pooled HRs was significantly and consistently $>1$ in subgroup meta-analysis stratified by the histology type and sample size. Furthermore, PCAT-1 high expression was an unfavorable independent prognostic factor for OS based on multivariate analysis (HR $= 1.04$, 95% CI: 1.01–1.07, $P < .001$).

3.3.3. Relationship between IncRNA PCAT-1 and clinico-pathologic features.

Pooled odds ratio (OR) for lncRNA PCAT-1 expression, presented in Table 3, showed that high expression of IncRNA PCAT-1 significantly correlated with depth of tumor invasion (OR $= 4.46$, 95% CI: 3.00–6.63, $P < .00001$), lymph node metastasis (OR $= 3.76$, 95% CI: 1.39–10.16, $P = .009$), and tumor stage (OR $= 4.09$, 95% CI: 1.35–10.82, $P = .004$). However, PCAT-1 expression was not associated with gender (OR $= 0.83$, 95% CI: 0.60–1.15, $P = .26$), differentiation (OR $= 1.20$, 95% CI: 0.78–1.83, $P = .40$), or distant metastasis (OR $= 1.49$, 95% CI: 0.70–3.15, $P = .30$).

3.3.4. Publication bias.

Begg test was used to assess the publication bias. Begg funnel plot with pseudo 95% CI was provided (Fig. 3). No significant publication bias affected the analysis of OS ($Pr = .188$).

3.3.5. Sensitivity analysis.

As illustrated in Figure 4, the result for sensitivity analysis for OS was negative, revealing that our results were relatively robust.

4. Discussion

The PCAT-1 is located in the chromosome 8q30 gene desert approximately 725 kb upstream of the c-MYC oncogene.$^{[14]}$ As a new identified prostate-cancer-associated lncRNA transcript 1, it was firstly reported to be implicated in prostate cancer progression and contributed to cell proliferation in prostate
Figure 2. Forest plot of the relationships between prostate-cancer-associated ncRNA transcript 1 (PCAT-1) and overall survival (OS).

Table 2
Results of subgroup analysis of pooled HRs of OS of cancer patients with high PCAT-1 expression.

| Stratified analysis                  | No. of Studies | No. of patients | Pooled HR (95% CI) | P   | I² (%) | P_h  |
|-------------------------------------|----------------|-----------------|--------------------|-----|--------|------|
| Cancer type                         |                |                 |                    |     |        |      |
| GC                                  | 2[24,25]       | 285             | 1.05 (1.02–1.08)   | <.001 | 35.5   | .213 |
| ESCC                                | 2[26,27]       | 451             | 1.04 (1.01–1.06)   | <.001 | 70.5   | .066 |
| Histology type                      |                |                 |                    |     |        |      |
| Squamous cell carcinoma             | 2[26,27]       | 451             | 1.04 (1.01–1.06)   | <.001 | 70.5   | .066 |
| Nonsquamous cell carcinoma          | 4[24,25,26,29] | 510             | 1.05 (1.02–1.08)   | <.001 | 36.5   | .193 |
| Sample size                         |                |                 |                    |     |        |      |
| ≥120                                | 3[25–27]       | 626             | 1.04 (1.01–1.07)   | <.001 | 59.7   | .084 |
| <120                                | 3[24,26,29]    | 535             | 1.05 (1.02–1.08)   | <.001 | 37.0   | .204 |
| Analysis type                       |                |                 |                    |     |        |      |
| Multivariate analysis               | 5[25–29]       | 851             | 1.04 (1.01–1.07)   | <.001 | 51.0   | .086 |

95% CI = 95% confidence interval, ESCC = esophageal squamous cell carcinoma, GC = gastric cancer, HR = hazard ratio.

Table 3
Results of meta-analysis of high PCAT-1 and clinicopathologic features.

| Category                                | Studies (n) | No. of patients | OR (95% CI) | P   | I² (%) | P_h  | Model          |
|-----------------------------------------|-------------|-----------------|-------------|-----|--------|------|---------------|
| Gender (male vs female)                 | 5[24,25,27–29] | 640             | 0.83 (0.60–1.19) | .26 | 0      | .43  | Fixed effects |
| Differentiation (poorly/undifferentiated vs well + moderately) | 3[25,26,29] | 400             | 1.20 (0.78–1.83) | .40 | 0      | .37  | Fixed effects |
| Tumor depth (T3–4 vs Tis–2)             | 4[24,25,27,29] | 523             | 4.46 (3.00–6.63) | <.0001 | 44     | .15  | Fixed effects |
| Lymph node metastasis (+ vs –)          | 4[24,25,27,29] | 523             | 3.76 (1.39–10.16) | .009 | 85     | .0002| Random effects |
| Distant metastasis (+ vs –)             | 5[24,25,27–29] | 630             | 1.49 (0.70–3.15) | .30 | 57     | .05  | Random effects |
| Tumor stage (III–IV vs 0–II)            | 4[24,25,27,29] | 532             | 4.09 (1.55–10.82) | .004 | 82     | .001 | Random effects |

95% CI = 95% confidence interval, OR = odds ratio.
In recent years, PCAT-1 has attracted great interest as a result of proof revealing that its abnormal expression in gastrointestinal cancer, such as HCC,[22,32,33] CRC,[21,23,29] GC,[24,25] ESCC,[26] and cholangiocarcinoma.[34] PCAT-1 was considered an oncogenic lncRNA in gastrointestinal tumors, and its overexpression was related to tumorigenesis and progression in various kinds of gastrointestinal cancers. PCAT-1 suppression significantly weakened cell proliferation, migration, and tumor invasion, whereas overexpressing PCAT-1 promoted these biologically aggressive features. Moreover, PCAT-1 could function as a competing endogenous RNA (ceRNA) to contribute tumor progression via several signaling pathways.

Figure 3. Funnel plots for publication bias test for overall survival (OS).

Figure 4. Sensitivity analysis for overall survival (OS).[24–29]
pathway, such as TP53-miR-215-PCAT-1-CRKL axis,[33] PCAT-1/miR-129-5p/HMGB1,[22] and PCAT1/miR-122/WNT1 axis.[14]

As far as we know, this is the first meta-analysis to comprehensively assess the association of PCAT-1 expression with prognosis and clinicopathologic features in gastrointestinal tumors. A total of 6 qualified studies, comprising 961 cases, were enrolled in this study. The pooled results showed that high expression of PCAT-1 was significantly associated with poor OS in gastrointestinal tumors, the subgroup analysis of PCAT-1 for OS further suggested that PCAT-1 could act as a predictive marker for OS in patients with gastrointestinal cancers. We also found that PCAT-1 was significantly correlated with some clinical features regarding tumor invasion, lymph node metastasis, and tumor stage. However, no obvious relationships were noticed between the high PCAT-1 expression and gender or differentiation or distant metastasis.

Some limitations of this study should be taken into account. First of all, the number of studies and the sample size were relatively small, with only 6 eligible studies with 961 cases were included. And then, all included studies were from China, researches from other countries were none or less, this may impact the broader applicability of our conclusions. Furthermore, there was no consensus on the cut-off value for distinguishing high and low PCAT-1 expression, for it was not easy to get a united threshold value in different studies. However, it is still essential to get a standardized value for PCAT-1 before it could be really applied in clinical practice. Additionally, significant heterogeneity was observed in the analysis of some clinicopathologic features. At last, most studies tended to report positive results rather than negative results, which might cause potential publication bias.

In conclusion, even with the limitations mentioned above, it can be preliminarily concluded that PCAT-1 might serve as a promising biomarker for improving prognosis estimation in gastrointestinal cancers. Notwithstanding, in the future, well-designed multicenter studies with large sample size are warranted to verify and strengthen the prognosis value of PCAT-1 in gastrointestinal cancers.

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References
[1] Torre LA, Bray F, Siegel RL, et al. Global cancer statistics, 2012. CA Cancer J Clin 2015;65:87–108.
[2] Chen W, Zheng R, Zeng H, et al. The incidence and mortality of major cancers in China, 2012. Chin J Cancer 2016;35:73.
[3] Lian D, Amin B, Du D, et al. Enhanced expression of the long non-coding RNA SNHG16 contributes to gastric cancer progression and metastasis. Cancer Biomark 2017;21:151–60.
[4] Zhang Y, Jin X, Wang Z, et al. Downregulation of SNHG1 suppresses cell proliferation and invasion by regulating Notch signaling pathway in esophageal squamous cell cancer. Cancer Biomark 2017;21:89–96.
[5] Zhu X, Chen F, Shao Y, et al. Long intergenic non-protein coding RNA 1006 was used as a potential novel biomarker of gastric cancer. Cancer Biomark 2017;21:73–80.
[6] Hu X, Sood AK, Dang CV, et al. The role of long noncoding RNAs in cancer: the dark matter matters. Curr Open Genet Dev 2017;48:8–15.
[7] Janardra A, Krause HM. The new RNA world: growing evidence for long noncoding RNA functionality. Trends Genet 2017;33:665–76.
[8] Pan P, Su W, Zhou Y. Precise long non-coding RNA modulation in visual maintenance and impairment. J Med Genet 2017;54:450–9.
[9] Gou X, Zhao X, Wang Z. Long noncoding RNA PVT1 promotes hepatocellular carcinoma progression through regulating miR-214. Cancer Biomark 2017;20:511–9.
[10] Sun X, Wang Z, Yuan W. Down-regulated long non-coding RNA SNHG1 inhibits tumor genesis of colorectal carcinoma. Cancer Biomark 2017;20:67–73.
[11] Zhang Y, Kong Z, Zhang Y. Increased expression of long non-coding RNA GLIDR in prostate cancer. Cancer Biomark 2017;19:145–50.
[12] Liu FT, Qu C, Luo HL, et al. The association of HOTAIR expression with clinicopathological features and prognosis in gastric cancer patients. Panminerva Med 2016;58:167–74.
[13] Liu FT, Dong Q, Gao H, et al. The prognostic significance of UCA1 for predicting clinical outcome in patients with digestive system malignancies. Oncotarget 2017;8:40620–32.
[14] Chandra Gupta S, Nandan Tripathi Y. Potential of long non-coding RNAs in cancer patients: from biomarkers to therapeutic targets. Int J Cancer 2017;140:1955–67.
[15] Liu F, Dong Q, Huang J. Overexpression of IncRNA PVT1 predicts advanced clinicopathologic features and serves as an unfavorable risk factor for survival of patients with gastrointestinal cancers. Cell Physiol Biochem 2017;43:1077–89.
[16] Reissner JR, Iyer MK, Balbin OA, et al. Transcriptome sequencing across a prostate cancer cohort identifies PCAT-1, an unannotated lincRNA implicated in disease progression. Nat Biotechnol 2011;29:742–9.
[17] Sarrafzadeh S, Geranpayeh L, Ghabouri-Fard S. Expression analysis of long non-coding PCAT-1 breast cancer. Int J Hematol Oncol Stem Cell Res 2017;11:185–91.
[18] Liu L, Liu Y, Zhuang C, et al. Inducing cell growth arrest and apoptosis by silencing long non-coding RNA PCAT-1 in human bladder cancer. Tumour Biol 2015;36:7685–9.
[19] Balci T, Yilmaz Susluer S, Kayabasi C, et al. Analysis of dysregulated long non-coding RNA expressions in glioblastoma cells. Gene 2016;590:120–2.
[20] Zhao B, Hou X, Zhan H. Long non-coding RNA PCAT-1 overexpression promotes proliferation and metastasis in non-small cell lung cancer cells. Int J Clin Exp Med 2015;8:14842–7.
[21] Qiao L, Liu X, Tang Y, et al. Down regulation of the long non-coding RNA PCAT-1 induced growth arrest and apoptosis of colorectal cancer cells. Life Sci 2017;188:37–44.
[22] Zhang D, Cao J, Zhong Q, et al. Long noncoding RNA PCAT-1 promotes invasion and metastasis via the miR-129-5p-HMGB1 signaling pathway in hepatocellular carcinoma. Biomed Pharmacother 2017;95:1187–93.
[23] Qiao L, Liu X, Tang Y, et al. Knockdown of long non-coding RNA prostate cancer-associated ncRNA transcript 1 inhibits multidrug resistance and c-Myc-dependent aggressiveness in colorectal cancer Caco-2 and HT-29 cells. Mol Cell Biochem 2018;441:99–108.
[24] Bi M, Yu H, Huang B, et al. Long non-coding RNA PCAT-1 overexpression promotes proliferation and metastasis in gastric cancer cells through regulating CDKN1A. Gene 2017;626:337–43.
[25] Cui WC, Wu YF, Qu HM. Up-regulation of long non-coding RNA PCAT-1 correlates with tumor progression and poor prognosis in gastric cancer. Eur Rev Med Pharmacol Sci 2017;21:501–7.
[26] Qin HD, Liao XY, Chen YB, et al. Genomic characterization of esophageal squamous cell carcinoma reveals critical genes underlying tumorigenesis and poor prognosis. Am J Hum Genet 2016;98:709–27.
[27] Bi M, Yu H, Li Q, et al. Upregulation of the long noncoding RNA PCAT-1 correlates with advanced clinical stage and poor prognosis in esophageal squamous carcinoma. Tumour Biol 2015;36:2501–7.
[28] Yan TH, Yang H, Jiang JH, et al. Prognostic significance of long non-coding RNA PCAT-1 expression in human hepatocellular carcinoma. Int J Clin Exp Pathol 2015;8:4126–31.
[29] Ge X, Chen Y, Liao X, et al. Overexpression of long noncoding RNA PCAT-1 is a novel biomarker of poor prognosis in patients with colorectal cancer. Med Oncol 2013;30:588.
[30] Prensner JR, Chen W, Iyer MK, et al. PCAT-1, a long noncoding RNA, regulates BRCA2 and controls homologous recombination in cancer. Cancer Res 2014;74:1651–60.
[31] Prensner JR, Chen W, Han S, et al. The long non-coding RNA PCAT-1 promotes prostate cancer cell proliferation through cMyc. Neoplasia 2014;16:900–8.
[32] Wen J, Xu J, Sun Q, et al. Upregulation of long non-coding RNA PCAT-1 contributes to cell proliferation, migration and apoptosis in hepatocellular carcinoma. Mol Med Rep 2016;13:4481–6.
[33] Ren Y, Shang J, Li J, et al. The long noncoding RNA PCAT-1 links the microRNA miR-213 to oncogene CRKL-mediated signaling in hepatocellular carcinoma. J Biol Chem 2017;292:17939–49.
[34] Zhang F, Wan M, Xu Y, et al. Long noncoding RNA PCAT1 regulates extrahepatic cholangiocarcinoma progression via the Wnt-catenin-signaling pathway. Biomed Pharmacother 2017;94:55–62.