Peer Review File

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Reviewer A

CSCCs in stage III cannot have curative surgery. Tuoheti et al (2020) has shown multiple generic variants about which the authors did not discuss. Finding have not shown in multivariate or otherwise that RIPK1 could be of much help over present prognostic criteria those author's mention.
Reply A: Because the patient indicated stage I or II before surgery, but the postoperative pathology suggested stage III.

Reviewer B

(1) There have been many studies on CSCC. What is the difference between this study and previous studies? What is the innovation? These need to be described in the introduction.
Reply B1: A large number of studies have confirmed that RIPK1 is related to the occurrence and development of tumors. This study identified high expression of RIPK1 in cervical squamous carcinoma and confirmed its clinicopathology.

(2) In the introduction of the manuscript, it is necessary to clearly indicate the relationship between RIPK1 and tumor-infiltrating immune cells and the role of RIPK1 play in prognosis in CSCC.
Reply B2: In this paper, we mentioned the role of RIPK1 in the prognosis of CSCC, but we did not verify the relationship between RIPK1 and tumor-infiltrating immune cells, and the review of the literature did not find the correlation between RIPK1 and tumor-infiltrating immune cells.

(3) In addition to RIPK1, what other genes play an important role in CSCC? It is recommended to add relevant content to the discussion.
Reply B3: There will be many genes that function in CSCC, but in this paper we only discuss the role of RIPK1 in CSCC.

(4) There are many genes that regulate CSCC. Why did the author choose RIPK1 for research? Please describe the reason. There are many genes that regulate CSCC. Why did the author choose RIPK1 for research? Please describe the reason.
Reply B4: Because RIPK 1 is associated with necrotic apoptosis of tumor cells, it is a tumor necrosis factor.

(5) Suggest adding functional research on RIPK1, which may be more meaningful.
Reply B5: We have partially addressed the function of RIPK 1 in the Discussion section.

(6) The introduction part of this paper is not comprehensive enough, and the similar papers have not been cited, such as "Construction and validation of prognostic prediction established on N6-methyladenosine related genes in cervical squamous cell carcinoma, Transl Cancer Res, PMID: 36237271". It is recommended to quote this article.
Reply B6: We read this article and found that it was similar to the one we cited, so considering the integrity of the article, it was more appropriate to choose the original literature.

(7) What is the role of RIPK1 in drug resistance in CSCC? Can RIPK1 become a therapeutic target for reversing drug resistance? Suggest adding relevant content.
Reply B7: Since we did not conduct relevant experiments on the functional role and therapeutic targets of RIPK 1 in CSCC, the relevant content was not added for the authenticity of the article.

Reviewer C

1) First, the title needs to indicate the clinical research design of this study, i.e., a retrospective cohort study.

Reply C1: We have modified our text as advised. (See page 1, line 4).

2) Second, the abstract needs some revisions. The background did not indicate the knowledge gap on the prognostic role of RIPK1 in CSCC and the potential clinical significance of this research focus. The methods need to describe the inclusion of subjects, the assessment of baseline clinical factors, follow up procedures, and measurements of prognosis outcomes. The results need to summarize the clinical characteristics of the study sample and quantify the findings by reporting statistics including expression levels, correlation coefficients, HR and P values. The conclusion should not mention "biological target for the treatment of CSCC" because the current study focuses on prognostic role.

Reply C2: We have modified our text as advised.

3) Third, in the introduction of the main text, the authors need to review known prognostic biomarkers in CSCC and have comments on their limitations and knowledge gaps. The potential strengths of RIPK1 in comparison to other known biomarkers should be described; otherwise, it remains unclear why RIPK1 deserves to be studied.

Reply C3: We have covered it in the Introduction section.

4) Fourth, in the methodology of the main text, please describe the clinical research design and details of the sample size estimation. In statistics, please describe the details of the multiple Cox regression analysis on the independent prognostic role of RIPK1, in particular how the clinical covariates were adjusted. The analysis on risk factors for the impaired prognosis of CSCC is not the focus of this study, the authors need to explain the reasons for this analysis. Please ensure P<0.05 is two-sided.

Reply C4: Multiple Cox regression analysis was performed to analyze whether RIPK 1 was an independent prognostic factor for CSCC. We confirm that P<0.05 is two-sided.

Reviewer D

1. Schmidt SV et al https://doi.org/10.18632%2Foncotarget.3249 did study the expression of RIPK in cervical SCC and your statement in line 75 is not valid. You can rephrase that statement after reading her publication.

Reply D1: We confirm the reading of her publication.

2. This a retrospective study and I suggest that in the "specimen characteristics" line 133 should read ...FFPE tissue blocks were retrieved for subsequent immunohistochemistry... The way it is written currently suggests you got fresh tissue samples and went through tissue processing? or was this the correct situation?

Reply D2: Because the sections we used were all early wax blocks, not fresh tissue samples.

3. Did you see the work of Ermine K et al https://doi.org/10.1016/j.gendis.2021.10.007? Do you think the findings in this paper could offer support to findings in the clinicopathologic characteristics?

Reply D3: We confirm that we see and believe that the finding can be supportive.

4. Line 163: "all the results were interpreted by a professional expert in pathology" is this expert a pathologist? if so, why not just state so? Is the pathologist part of the study?

Reply D4: this expert a pathologist.

5. In examining the IHC slides was a conventional light microscope used or digital scanned slides were used in assessing the slides? If a microscope was used details of the microscope and its field size will be very important in standardizing the grading of the staining.

Reply D5: We used a conventional light microscope.

6. Image 1E. Kindly label the details that you intend to show to the readers.

Reply D6: This figure is designed to show the picture at different multiples in the same position.

7. Figure 2. Correct the spelling of "cell carcinoma"

Reply D7: We have modified our text as advised. (See page 12, line 360-361).

Reviewer E

1. Reporting Checklist

Please provide the section and paragraph in the checklist for items in green box. And Please heck if item 18 is applicable, if not, please fill N/A.

RESULTS			
Data			
12	Describe the flow of patients through the study, including the number of patients included in each stage of the analysis (a diagram may be helpful) and reasons for dropout. Specifically, both overall and for each subgroup extensively examined report the numbers of patients and the number of events.	8/155	Patient pathological characteristics
13	Report distributions of basic demographic characteristics (at least age and sex), standard (disease-specific) prognostic variables, and tumor marker, including numbers of missing values.	8/162	expression of RIPK1 in CSCC and normal samples
Analysis and presentation			
14	Show the relation of the marker to standard prognostic variables.	8/155	Results
15	Present univariable analyses showing the relation between the marker and outcome, with the estimated effect (e.g., hazard ratio and survival probability). Preferably provide similar analyses for all other variables being analyzed. For the effect of a tumor marker on a time-to-event outcome, a Kaplan-Meier plot is recommended.	8/172	Results
16	For key multivariable analyses, report estimated effects (e.g., hazard ratio) with confidence intervals for the marker and, at least for the final model, all other variables in the model.	9/185	results
17	Among reported results, provide estimated effects with confidence intervals from an analysis in which the marker and standard prognostic variables are included, regardless of their statistical significance.	9/179	results
18	If done, report results of further investigations, such as checking assumptions, sensitivity analyses, and internal validation.		
DISCUSSION			
19	Interpret the results in the context of the pre-specified hypotheses and other relevant studies; include a discussion of limitations of the study.	11/238	Discussion
20	Discuss implications for future research and clinical value.	12/242	discussion

Reply:we have added as advised.(See checklist)

2. Figure 1

Please explain IHC in the legend.

Reply:we have added as advised.(See page 12,line 372)

3. Table 2

Please add the description to the table footnote that how the data are presented in table.

FIGO stage				41.830	0.007**			36.369	0.028*
IA+IB	51	32±5	30–34			32±4	31–34		
Ⅱ+III	49	30±7	29–32			31±7	29–32		
Histological grade				33.295	0.058			24.869	0.303
G1/G2	79	31±6	29–32			31±6	30–33		
G3	21	30±6	27–33			30±6	28–33		
Lymph node metastasis				40.375	0.001**			31.293	0.090

Reply:we have added as advised.(See table 2 footnote)

4. References/Citations

a) If available, please update your reference list by including related literatures published in 2022. Some of the references are outdated.

Reply:We believe that the references cited are necessary for their existence.

b) Please double-check if citations should be added as you mentioned “studies”.
*Please note that the references should be cited in order of their appearance in the text. If the studies are not included in the reference list, please also update the current version.

263 cervical cancer and has high mortality rates, and epidemiology studies have reported
264 the age of onset in patients with CSCC is decreasing worldwide. CSCC are prone to
265 develop tumor metastasis and have a high rate of recurrence. Thus, the present study

Reply:we have modified our text as advised.(See page 8,line 240)

c) Please double-check if more studies should be cited as you mentioned “studies”. OR use “study” rather than “studies”.

273 and FLIP (and also caspase 10 in humans). Previous studies have shown that
274 complexes that contain necrosomes, such as RIPK1 and/or RIPK3, can activate
275 caspase 8 and induce apoptosis (15). However, RIPK1 kinase activity-related cell

Reply:we have modified our text as advised.(See page 8,line 250)