# Peer Review File

Article information:

This is an interesting study with potential clinical relevance. There are a few, relatively minor issues to address to improve the quality of the study / manuscript.

1.Lines 277-279: On the other hand, the high-risk group had a higher proportion of activated CD4 T cells, CD56 natural killer cells, γδ T cells, natural killer T cells, neutrophils, and Th2 cells than the low-risk group (Figure 9A). – the results seem counterintuitive. Any explanation?

**Reply 1:** We sincerely appreciate the valuable comments. Here, some factors may need to be considered, such as the complexity and diversity of the immune system. Although the high-risk group shows more active specific immune cell types, it does not necessarily indicate a good immune response. Sometimes, over-activated immune cells can lead to an imbalance in the immune system, which negatively affects anti-tumor effects. At the same time, the high-risk group displays a greater number of immune cells, but this does not necessarily mean that these immune cells' functions are normal. In the future, we will further explore the specific roles among immune cells in the high-risk group.

2.Figure 5H AUC at 1 year, 3 year and 5 year (described as AUC at 1 year, 2 year and 3 year in the graph) in the external validation cohort were not as significant as I would have hoped. Any explanation?

**Reply 2:** We sincerely appreciate the valuable comments. We have re-uploaded the images of AUC at 1 year, 3 years, and 5 years; however, the external validation cohort still did not reach a significantly high level. We believe that this discrepancy may be attributed to the differences in sequencing methods (patient sample chip sequencing for the GEO database and RNA-seq data for TCGA), resulting in variations in data types and introducing some errors. Additionally, the sample sizes of lung adenocarcinoma patients in the GEO and TCGA databases differ, which could also contribute to the lack of significance. In the future, we plan to include more clinical samples for further validation.

**Changes in the text: Figure 5H**

3.Not only the roles of ACTB, PDGFB and CPS1 on carcinogenesis, cancer invasion and metastasis, etc. but also those of other stemness-related genes, which were identified in this study, should be discussed appropriately.

**Reply 3:** We completely agree with your point of view. We have added it to the discussion section of the article.

**Changes in the text:** Page18-20, line 361-402.

4.Clinical characteristics evaluated with univariate Cox regression analysis only consisted of age, gender and stage. The authors discussed the lack of information about adverse histologic

factors in this analysis, but information about systemic therapies (adjuvant or first-line treatment) should also be included especially because the authors advocate that the prognostic risk model can help the clinician develop appropriate treatment plans for maximizing clinical benefits.

**Reply 4：** We completely agree with your comments. The information regarding systemic treatments (adjuvant or first-line therapy) received by patients is crucial for constructing a prognostic risk model. In this study, we were unable to account for this information, which has been acknowledged as a limitation in the discussion section. In the future, we will further investigate with the hope of developing a more comprehensive prognostic model.

**Changes in the text:** Page 17, line 329-330.

Additional issues:

1.Highlight Box, What is the implication… I don't understand the following sentence. Future diagnosis and treatment will further focus on Mechanism research.

**Reply 1:** We sincerely appreciate the valuable comments. We apologize for not accurately conveying the meaning of the sentence. we have modified our text as "Future diagnosis and treatment will further focus on stemness perspectives." The meaning of the sentence is that in the future, it will be possible to measure the expression of certain stemness-related genes that are strongly correlated with prognosis during diagnosis. By screening out patients with higher stemness, targeted treatments can be achieved.

**Changes in the text:** Page 4, line 52.

2.Line 276: What do columnar cells represent? They do not seem to comprise an immune cell subtype.

**Reply 2:** We greatly appreciate your careful identification of this error. We apologize for my carelessness in referring to mast cells as columnar cells in Figure 9A. we have modified our text.

**Changes in the text:** Page 15, line 282.

==Reviewer B==

1.  It is suggested to cite the author name in the following sentence since you mentioned the author name:
    -   This index model was established by ==Malta et al==. using machine learning algorithms based on a dataset of progenitor cells. This method allows for the calculation of mRNAsi for samples in the TCGA database through RNA-seq analysis, enabling the evaluation of their stem cell properties.

**Reply 1:** We agree with your assessment. we have modified our text as advised.

**Changes in the text:** Page6, line 86-89.

2.  Please check if more references should be cited in the following sentence since you mentioned "studies".
    -   ==Studies (4) have== confirmed that CSCs possess significant characteristics such

as self-renewal and multi-lineage differentiation, which can lead to tumor metastasis, drug resistance, and recurrence.
- Studies have shown that CPS1 is downregulated in hepatocellular carcinoma, and its low expression predicts poor prognosis for patients (26).
- Experimental studies (29) have shown that when EGFR is inhibited, lung adenocarcinoma cells become more dependent on the urea cycle, particularly CPS1. Inhibition of both CPS1 and EGFR suppresses cell cycle progression and cell proliferation.

**Reply 2:** We greatly appreciate your careful identification of this error. I sincerely apologize for the failure to distinguish between singular and plural, as all three sentences specifically refer to the study cited in this particular reference. And we have modified "studies" to "the study" in our text.

**Changes in the text:** Page 5, line 77; Page 18, line 413; Page 19, line 425.

3. Figures
   (1) The data are too close to distinguish in Figure 1A. please check and revise.
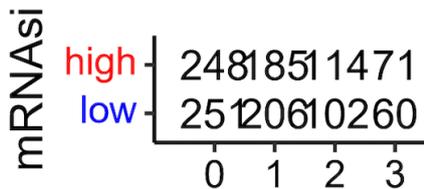


**Reply 3(1):** We sincerely appreciate the valuable comments. we have modified Figures 1A as advised.

**Changes in the text:** Figure 1A.

   (2) It is suggested to extend the x-axis to 2.0 in Figures 5E-5F.

**Reply 3(2):** We sincerely appreciate the valuable comments. we have modified Figures 5E-5F as advised.

**Changes in the text:** Figures 5E-5F.

   (3) Please revise "riskScore" to "Risk Score" in Figures 5E-5F.

**Reply 3(3):** We sincerely appreciate the valuable comments. we have modified Figures 5E-5F as advised.

**Changes in the text:** Figures 5E-5F.

   (4) Figure 5E-F: Please revise "pvalue" to "P value" and "Hazard ratio" to "Hazard ratio (95%CI)".



**Reply 3(4):** We sincerely appreciate the valuable comments. we have modified Figures 5E-5F as advised.

**Changes in the text:** Figures 5E-5F.

(5) "risk socre" should be "risk score" in Figures 6B-6C, 7C-7F.

Patients (increasing risk <u>socre</u>)

**Reply 3(5):** We sincerely appreciate the valuable comments. we have modified Figures 6B-6C, 7C-7F as advised.
**Changes in the text:** Figures 6B-6C, 7C-7F.

(6) Please indicate the meaning of "***" in Figure 8A.
**Reply 3(6):** We sincerely appreciate the valuable comments. "***" in Figure 8A means P<0.001.
**Changes in the text:** Page32, line 709.

(7) Please indicate the full name of "MNA" in Figure 8 legend.

Points

0

MNA

**Reply 3(7):** We sincerely appreciate the valuable comments. We have modified "MNA" to "M((NA)"in Figure 8A legend. M(NA) means no data for M staging.
**Changes in the text:** Page32, line 709-710.

(8) Please add a unit to futime in Figure 8A.
**Reply 3(8):** We sincerely appreciate the valuable comments. We have modified Figures 8A as advised.
**Changes in the text:** Figure 8A

(9) Please delete "(%)" in the axes of Figure 8B since their rates are 0-1.
**Reply 3(9):** We sincerely appreciate the valuable comments. we have modified Figures 8B as advised.
**Changes in the text:** Figure 8B