Peer Review File

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Reviewer A

1. The study searched the TCGA database for DEGs and clinicopathological information. The clarity on the criteria for selecting the 502 LSCC patients and 49 normal adjacent lung tissues is crucial.
Reply 1: We appreciate the reviewer's careful review of our manuscript. 502 LSCC patients and 49 normal adjacent lung tissues were the data of all LSCC in the Cancer Genome Atlas (TCGA) database up to November 15, 2022.
Changes in the text: None

2. The inclusion of 29 m7G methylation-related genes in constructing the risk model is used. However, transparency on the rationale for selecting these specific genes is necessary. The impact of the chosen genes on the risk model and their relevance to LSCC should be explicitly justified.
Reply 2: We agree with the reviewer's comments. 29 m7G methylation-related genes were all genes in publicly available data up to the time we were ready to study. Based on the role of these genes, we explored their potential to predict the prognosis of LSCC.
Changes in the text: None

3. While the study used limma R package for differential expression analysis, the justification for selecting a log fold change (FC) >0.5 and an adjusted P value <0.001 as criteria for defining DEGs should be provided. These thresholds can significantly affect the identification of DEGs.
Reply 3: Thank you for pointing this out, this value is set according to the empirical value, and the more demanding P-value enables better DEGs.
Changes in the text: None

4. The authors should provide a clear explanation of the survival analysis methodology, including the specific statistical tests used in univariate Cox regression, is crucial. The significance threshold for selecting genes and the subsequent construction of the risk prediction model needs to be justified also.
Reply 4: Thank you for pointing this out, which we had not fully acknowledged in our original manuscript. we added "Statistical Analysis" in "Method" section.
Changes in the text: see the Page 6; line 171-176.

5. While constructing the PPI network, the authors should clarity on how the biological relationships were determined and the criteria for indicating upregulated and downregulated correlations is needed. The interpretation of the network's biological significance should be explicitly addressed.
Reply 5: We agree with the reviewer's comments.We have revised the manuscript according to the reviewer's comment.

Changes in the text: see the Page 10, line 315-320.

6. The study used GO and KEGG pathway analyses, but the specific processes and criteria used for categorizing patients into high- or low-risk categories for subsequent analyses need to be detailed. The rationale for choosing the median risk score as the cut-off should be explained.
Reply 6: We agree with the reviewer's comments. We calculated the risk score of each patient according to the risk model formula (see the Page 7, line 211-213), and then took the median value of all scores as the cut-off value to divide the patients into high- and low-risk groups. In risk assessment, especially when extreme values or outliers are present, the use of medians can reduce the impact of these extreme values on the overall score, thus providing a more accurate and reliable risk assessment result.
Changes in the text: None

7. The methodology for profiling immune cell populations, immune-associated pathways, and the role of gene set signatures in LSCC should be clarified. The specific signatures used and the thresholds for determining enrichment scores need explicit explanation.
Reply 7: We agree with the reviewer's comments. As recommended, we have revised the manuscript according to the reviewer's comment.
Changes in the text: see the Page 10, line 324-329.

8. The study should address how missing data, if any, was handled during the analysis. Missing data can introduce bias and impact the validity of results.
Reply 8: Thank you for pointing this out, the data used for analysis has been cleaned, so there is no missing data.
Changes in the text: None

9. More references on bioinformatics workflow should be added to attract a broader readership i.e., PMID: 36936815, PMID: 35851932.
Reply 9: We have added the references to the revised manuscript
Changes in the text: see the Page 9, line 274-275.

10. A more comprehensive review of current treatment strategies would add depth to the context.
Reply 10: We agree with the reviewer's comments. We have revised the manuscript according to the reviewer's comment.
Changes in the text: see the Page 9, line 274-275.

11. While the study emphasizes the need for novel biomarkers, it lacks a direct link between the identified m7G methylation-related genes and their potential as biomarkers. The importance of these genes in predicting treatment outcomes should be more explicitly justified.
Reply 11: We appreciate and agree with the reviewer's suggestion, because of objective reasons such as funding etc, we did not carry out verification in this study. At present, we are actively seeking funds to verify our conclusions.
Changes in the text: None

12. The claim that the risk model predicts overall survival (OS) needs stronger support.
Reply 12: We appreciate and agree with the reviewer's suggestion. To establish m7G methylation-related genes and their potential as biomarkers in future,we plan to build in vivo and in vitro experimental models to verify the therapeutic significance.
Changes in the text: None

13. The biological functions of the identified genes are briefly mentioned, but a deeper discussion on their roles in cancer, especially in lung cancer, is necessary.
Reply 13: We have revised the manuscript according to the reviewer’s comment.
Changes in the text: see the Page 10, line 315-329.

14. While stating that the study is one of the few exploring m7G methylation in lung cancer, a more detailed comparison with existing studies is needed. What distinguishes this study from previous ones, and how does it contribute to the existing knowledge?
Reply 14: We appreciate the reviewer’s encouragement and helpful comment. We have revised the manuscript according to the reviewer’s comment.
Changes in the text: see the Page 9, line 274-275 and Page 10, line 315-329.

15. The study lacks insight discussions into the functional mechanisms of these genes in LSCC development and progression. A more detailed exploration of the biological roles of these genes is required.
Reply 15: We thanks for reviewer’s suggestion. As we shown in the paper (see Page 11, line 335-336), it was our limitation, and also was what we're going to do next.
Changes in the text: None

16. Only data from the TCGA database was analyzed raises concerns about the lack of information on the protein-level expression of the identified genes. The importance of validating findings at the protein level for clinical relevance should be emphasized.
Reply 16: We strongly agree with the reviewer's suggestion. In this study, we explored the possibility of m7G methylation-related genes predicting the prognosis of LSCC, and built a prediction model. Due to the lack of funding, we have not been verified at the protein level , which is also one of our limitation (see Page 10, line 331-334) . In the future, we will strive for more funding to verify our results.
Changes in the text: None

17. Internal/external validation is crucial for confirming the generalizability of results.
Reply 17: We strongly agree with the reviewer's suggestion. In this study, we explored the possibility of m7G methylation-related genes predicting the prognosis of LSCC, and built a prediction model. Due to the lack of funding, we have not been verified, which is also one of our limitation (see Page 10, line 331-334 and Page 11, line 335-336) . In the future, we will strive for more funding to verify our results.
Changes in the text: None

Reviewer B

A signature of five 7-methylguanosine-related genes is a prognostic marker for lung squamous cell carcinoma
After carefully reviewing the manuscript, I find that it has significant potential for publication in JTD. However, I have a few minor recommendations that, if addressed, would further enhance the manuscript.

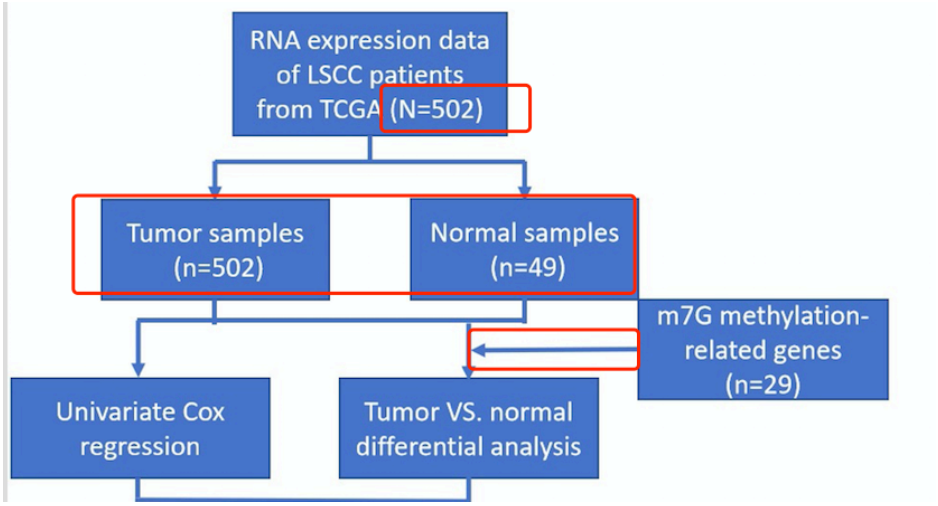
Comment 1: Please provide a more detailed discussion on the novelty of your findings and how they contribute to the existing knowledge in the field.
Reply 1: We have revised the manuscript according to the reviewer's comment.
Changes in the text: see the Page 10, line 315-329.

Comment 2: I recommend validating the performance of your predictive biomarkers in independent cohorts using public datasets or experimental qPCR. This will help assess the robustness and generalizability of your findings and provide further support for the potential clinical utility of the 7-methylguanosine-related gene signature in LUSC prognosis.
Reply 2: We strongly agree with the reviewer's suggestion. In future, we plan to build in vivo and in vitro experimental models to verify the therapeutic significance.
Changes in the text: None

Comment 3: To enhance the reproducibility of your study, please include a detailed description of the analysis pipelines used.
Reply 3: We have revised the manuscript according to the reviewer's comment.
Changes in the text: see the Figure 1.

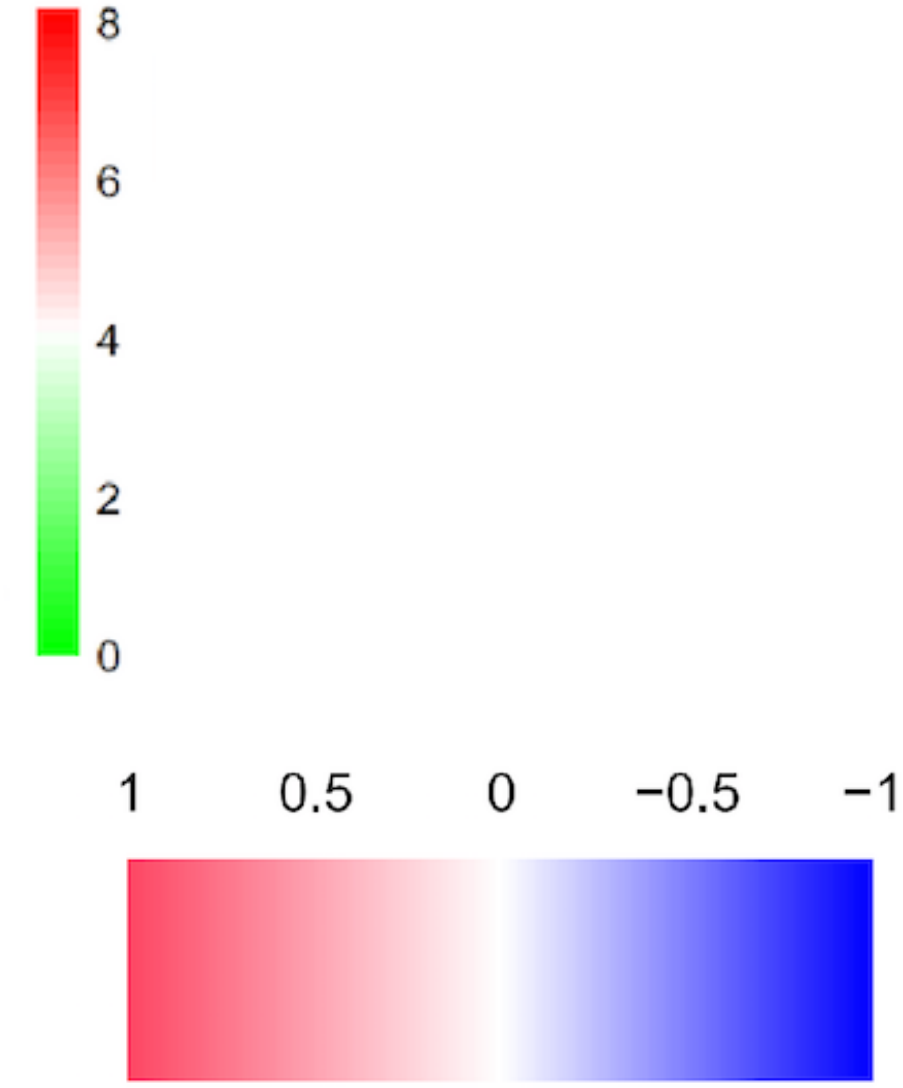
Reviewer C

1. Figure 1
Please check the data.



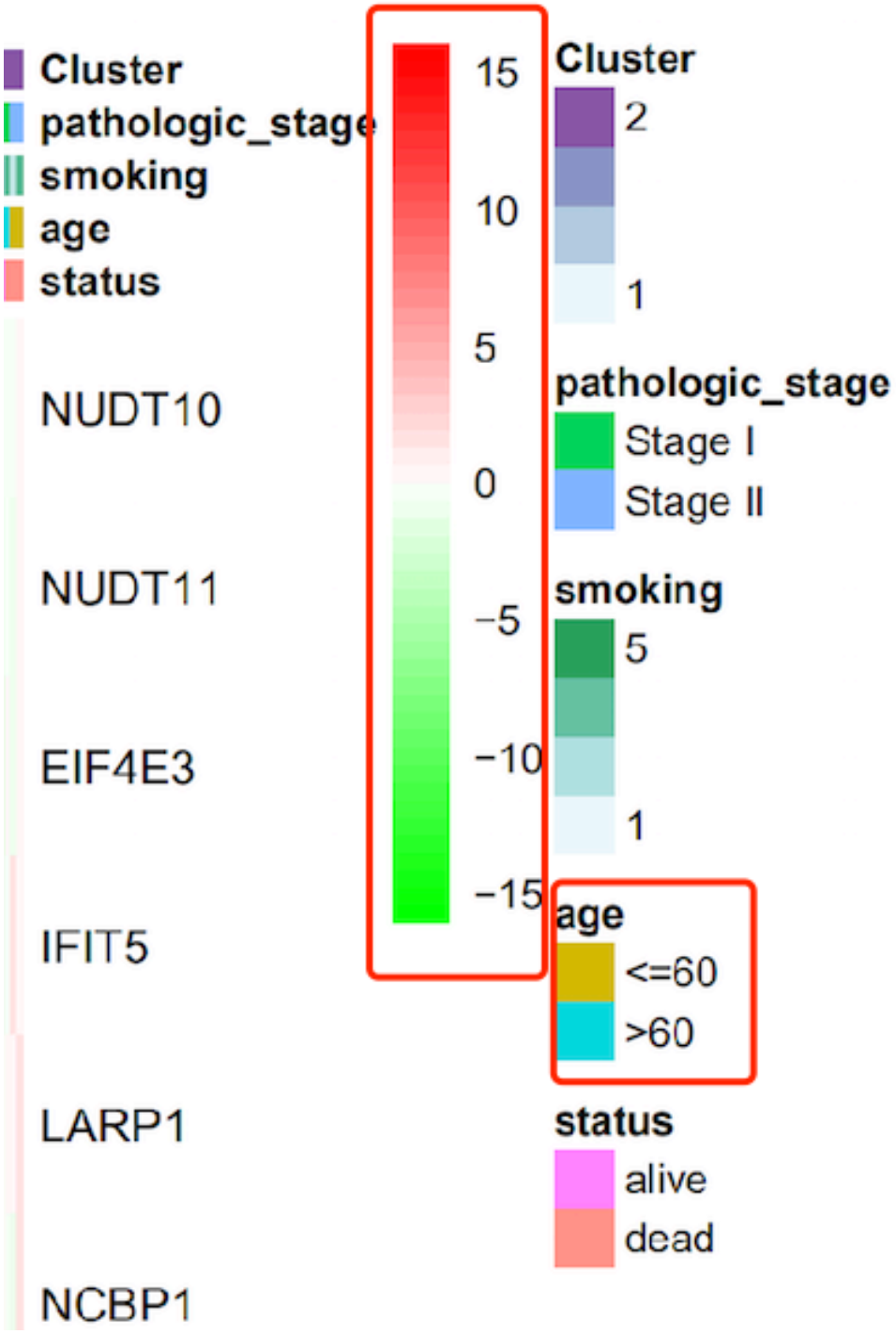
Rely: we have redrawn Figure 1.

2. Figure 2A & Figure 2B
Please explain the meaning of the below bar.



Rely: we have added this contents in Figure 2 legend in revised manuscript.

3. Figure 3B
1) Please explain the meaning of the below bar.
2) Please provide the unit for “Age”.

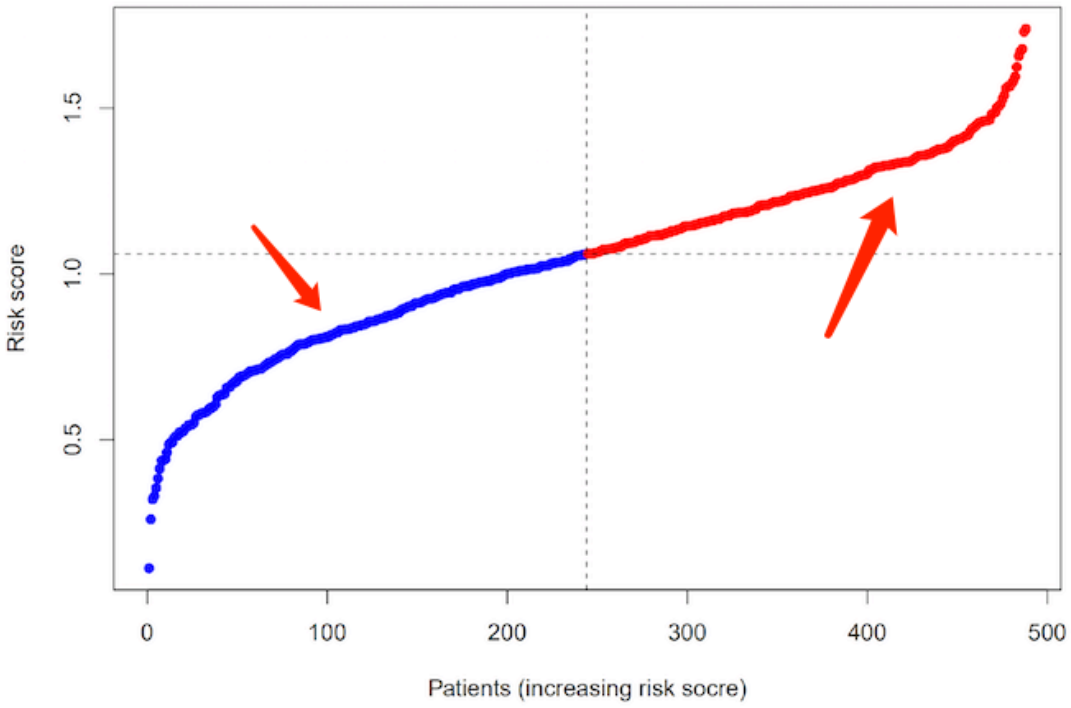


Rely: we have added this contents in Figure 3B and Figure 3B legend in revised manuscript.

4. Figure 4C

Which one means high? Which one means low? Please check and indicate.

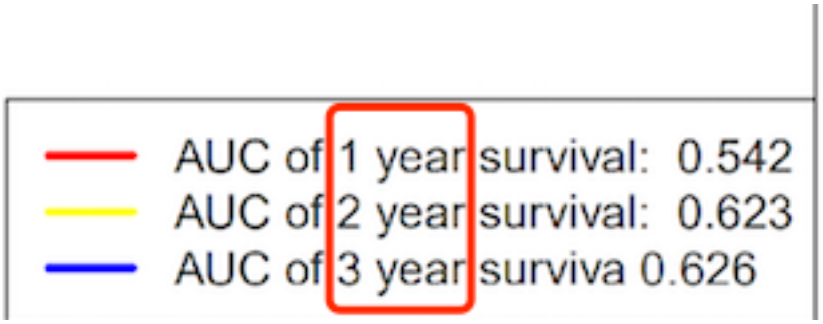
C



Rely: we have added this contents in Figure 4C in revised manuscript.

5. Figure 4F

Please change “1 year”, “2 year”, “3 year” to “1-year”, “2-year”, “3-year”. Please check and revise.

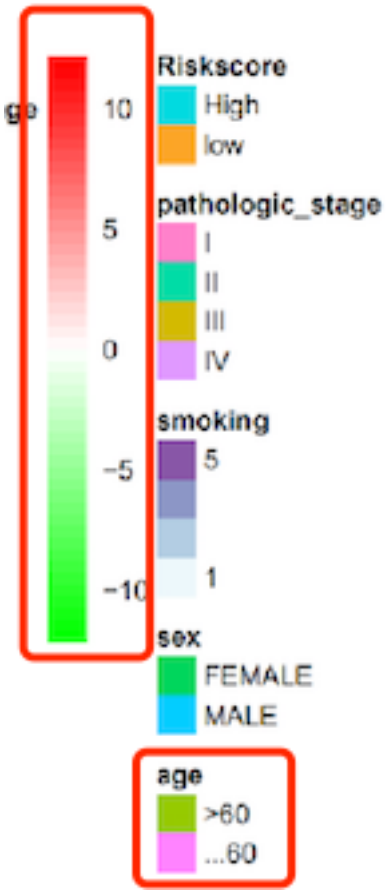


Rely: we have changed in Figure 4F in revised manuscript.

6. Figure 5C

1) Please explain the meaning of the below bar.

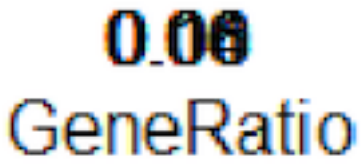
2) Please provide the unit for “Age”.



Rely: we have added this contents in Figure 5C and Figure 5c legend in revised manuscript.

7. Figure 6B

The data was covered. Please check and revise.



Rely: we have redrawn Figure 6.