

Ref.: NO.111171

Title: Tumour chemotherapy sensitivity test may predict clinical outcomes in colorectal cancer patients receiving oxaliplatin and fluoropyrimidine-based regimens

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Dear Editor,

Thank you very much for the comments on the manuscript (NO.111171) entitled "Tumour chemotherapy sensitivity test may predict clinical outcomes in colorectal cancer patients receiving oxaliplatin and fluoropyrimidine-based regimens", which was submitted to the "*World Journal of Gastrointestinal Oncology*". We appreciate the valuable suggestions. We have carefully read the reviewers' comments and revised the manuscript accordingly. Enclosed please find our point-by-point responses.

The revised sections in our manuscript, in accordance with the reviewers' suggestions, are highlighted in yellow. The manuscript has also been revised by a professional English language editing company. Please feel free to contact us if you have any questions or concerns.

Thank you again for your comments.

Yours sincerely,

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Point-by-point response

REVIEWER COMMENTS

Reviewer #1:

Your research work has conducted an in-depth assessment of the response of colorectal cancer patients to the combined chemotherapy regimen of fluorouracil and oxaliplatin through the ATP-TCA chemical sensitivity test. The research results clearly confirm that there is a significant correlation between the ATP-TCA chemical sensitivity test and the three-year and five-year overall survival and progression-free survival rates of resectable colorectal cancer patients after surgery combined with chemotherapy. This research not only provides a new perspective for the treatment of colorectal cancer, but also your findings have important guiding significance for clinical practice. Although your research has achieved remarkable results, there are still some aspects that may need further improvement and enhancement. For example, you constructed a Cox regression model in your research, which is an extremely important and widely used model in medical statistical analysis. However, the application of the Cox regression model in your research seems to need further refinement. Specifically, your model construction process did not include any internal or external validation steps. An unvalidated model, no matter how excellent its AUC (Area Under the Curve) and C-index (Concordance Index) values are in the training set, has limited practical significance. Moreover, the AUC and C-index values shown by the training set data you used are not particularly outstanding, which raises doubts about the generalization ability of the model and the reliability of the series of conclusions you have drawn based on the current model. An unvalidated Cox regression model may only be able to identify the correlation between parameters and clinical outcomes. Therefore, I strongly recommend that you add model validation steps in your subsequent research, which will greatly enhance the credibility and persuasiveness of your research results. I firmly believe that if your research can be further

optimized and improved in this aspect, it will not only significantly enhance the quality and depth of the research, but also gain wider recognition from the academic and clinical communities, thereby having a more profound impact on the research of colorectal cancer treatment.

Answer

Thank you for the comments.

As the expert rightly noted, a Cox regression model that has undergone both internal and external validation can more reliably determine the association between parameters and clinical outcomes. Based on this suggestion, we randomly divided the original data into a training set and a validation set, rebuilt the Cox regression model, and performed internal validation, thereby increasing the scientific rigor of the research findings. Although we are currently unable to obtain multicentre data for external validation, we thoroughly addressed the corresponding limitations and future research plans in the discussion section, aiming to objectively reflect the inferential boundaries of this study under current conditions and outline directions for further research. The relevant modifications and additions are highlighted in yellow in the manuscript.

Reviewer #2: This study conducted a retrospective analysis of ATP-TCA test results from 1,549 colorectal cancer patients, confirming the independent prognostic value of ATP-TCA in predicting overall survival (OS) and disease-free survival (DFS) among resectable colorectal cancer patients receiving oxaliplatin- and fluorouracil-based adjuvant chemotherapy regimens. The findings demonstrate that ATP-TCA can help optimize postoperative chemotherapy selection, enhance treatment efficacy, and improve patient outcomes. Although certain limitations exist, such as the retrospective design and inconsistent thresholds, the research provides important evidence for future developments in personalized medicine and recommends further large-scale prospective studies to validate its clinical

application value. The present investigation explores the predictive value of ATP-TCA testing in individualized chemotherapy for colorectal cancer, featuring a sufficient sample size and rigorous statistical analyses (Cox regression, Kaplan-Meier survival analysis, etc.), with clear clinical significance.

However, three limitations—sample selection bias, insufficient threshold standardization, and relatively short follow-up periods—require targeted improvements to enhance the reliability of the conclusions. The revision suggestions are as follows.

1. **Sample selection bias:** Excluding patients who received neoadjuvant/targeted/immunotherapy (proportion in sample unknown) limits the results to traditional chemotherapy Homo sapiens populations, deviating from modern clinical practice. Discuss the impact of exclusion criteria on the generalizability of conclusions and quantify the proportion of excluded patients in the "Limitations" section.

Answer to 1

We thank the reviewers for their valuable suggestions. Please refer to the yellow-highlighted sections in the Methods section, Figure 1 and the Discussion section for details.

2. **Standardization of ATP-TCA threshold:** The self-defined IC50/IC90 threshold lacks validation of consistency with thresholds from other studies (e.g., international consensus). Cite prior literature (e.g., ATP-TCA threshold studies in breast cancer) to justify the current threshold.

Answer to 2

We appreciate the reviewers' insightful suggestion. The relevant literature has been cited in the Methods section to provide a rationale for the ATP-TCA threshold standardization.

3. **Insufficient follow-up time:** A median follow-up of 2.78 years (33 months) is inadequate for evaluating long-term survival in early-stage colorectal cancer (stages I-II). Extend follow-up to ≥ 5 years (especially for

early-stage patients) and supplement with 5-year survival rate analysis. If follow-up extension is unfeasible, explicitly state in the discussion that "conclusions are limited to mid-term prognosis."

Answer to 3

We appreciate the reviewers' insightful suggestion. Given the current limited follow-up duration, we will continue to extend the study period to obtain longer-term data. Furthermore, the Discussion has been revised to explicitly state that the conclusions of this study are limited to midterm prognosis.

4. Clinical translation supplement: Discuss the predictive advantages/disadvantages of ATP-TCA compared to genetic testing (e.g., RAS mutation) and PD-L1 testing.

Answer to 4

We appreciate the reviewers' insightful suggestion. The Introduction section has been updated with a comparison of ATP-TCA to other individualized methods, such as genetic and PD-L1 testing.

5. Rigorous phrasing: For example, replace "fills the research utetheisa kong white" with "supplements evidence on ATP-TCA application in colorectal cancer." Clearly state study limitations (sample selection, follow-up duration) in the abstract/conclusion.

Answer to 5

We thank the reviewers for their valuable suggestions and have rigorously revised our conclusions and study limitations accordingly. Please refer to the Abstract section, Pg 2, and the Conclusion section for details.

6. Standardize table formats throughout the manuscript (e.g., table titles).

Answer to 6

Thank you for the comments. The formatting settings of all the tables have been standardized.