nature portfolio

Peer Review File

Characterizing mutation-treatment effects using clinico-genomics data of 78,287 patients with 20 types of cancers

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This manuscript has been previously reviewed at another journal. This document only contains information relating to versions considered at Nature Communications.

This file contains all reviewer reports in order by version, followed by all author rebuttals in order by version.

Version 0:

Reviewer comments:

Reviewer #4

(Remarks to the Author)
In this revised manuscript, the authors have tried to address the comments from the reviewers. The reviewer still has several remaining concerns about the background information/knowledge for lung cancer mutations and treatments. The reviewer thinks it important for understanding mutation-treatment relationships.

Point:
1. The authors conducted the analysis considering only known and likely pathogenic mutations according to the reviewer's previous comments. The reviewer could not find the definition of "known" and "pathogenic" mutations in the manuscript. The authors should carefully explain how the impact of mutations were determined.

2. To the reviewer's understandings, some kinds of TKI treatments are applied to patients with target mutations (e.g. osimertinib treatment for EGFR mutants). Did the patients with EGFR inhibitors have EGFR driver mutations? The author should describe such background information of each gene and treatment when they discussed the specific example.

(Remarks on code availability)
The reviewer checked the github page but did not run the codes.

Reviewer #5

(Remarks to the Author)
Comments to responses

While the authors have adequately addressed the concerns raised by reviewers #2 and #3, I share the concern pointed out by reviewer #2 regarding the novelty of the findings. The current manuscript does not appear to offer significantly new insights when compared to the authors' previous work. The additional predictive mutation-treatment interactions and the machine learning model for risk score prediction, which have limited clinical applicability or validation, may not provide sufficient impact to justify publication in Nature Communications. However, I would defer to the editor's judgment on this matter.

Further comments for consideration

Despite the authors have stated in their response to Reviewer #1 that "The current study did not include an analysis of co-mutations," it would be beneficial to include a simple analysis that could provide valuable insights. A basic visual representation, such as an oncoplot
(e.g., https://bioconductor.org/packages/release/bioc/vignettes/maftools/inst/doc/maftools.html#72_Oncoplots), could illustrate co-occurring or mutually exclusivity of predictive mutations across different cancer types. For instance, in a cohort of non-small cell lung carcinoma, patients could be stratified based on the co-occurrence or mutual exclusivity of TP53 and

CDKN2A/B mutations. The authors could then demonstrate the differences in the impact on treatment outcomes between these stratified groups.

Typo

line 135: gene-mutation -> gene-treatment

(Remarks on code availability)

REVIEWERS' COMMENTS

Reviewer #4 (Remarks to the Author):

In this revised manuscript, the authors have tried to address the comments from the reviewers. The reviewer still has several remaining concerns about the background information/knowledge for lung cancer mutations and treatments. The reviewer thinks it important for understanding mutation-treatment relationships.

Point:
1. The authors conducted the analysis considering only known and likely pathogenic mutations according to the reviewer's previous comments. The reviewer could not find the definition of "known" and "pathogenic" mutations in the manuscript. The authors should carefully explain how the impact of mutations were determined.

Thank you for this comment. We have added more clarifications of this in the Methods:

> Pathogenic variants, both known and likely, were identified using a tailored filtering approach[38]. Germline variant predictions were excluded through cross-referencing with publicly available databases, including the 1000 Genome Project and the Exome Aggregation Consortium (ExAC), while retaining established cancer-driving events. Additionally, likely germline variants of uncertain significance were excluded using an algorithm distinguishing somatic from germline variants without matched normal tissue[39]. For further validation, pathogenic variants were corroborated, and truncations and deletions in tumor suppressor genes were categorized as likely pathogenic.

References:

38. Hartmaier, R. J. *et al.* High-throughput genomic profiling of adult solid tumors reveals novel insights into cancer pathogenesis. *Cancer Research*, 77(9), 2464-2475 (2017).
39. Sun, J. X. *et al.* A computational approach to distinguish somatic vs. germline origin of genomic alterations from deep sequencing of cancer specimens without a matched normal. *PLoS Computational Biology*, 14(2), e1005965 (2018).

2. To the reviewer's understandings, some kinds of TKI treatments are applied to patients with target mutations (e.g. osimertinib treatment for EGFR mutants). Did the patients with EGFR inhibitors have EGFR driver mutations? The author should describe such background information of each gene and treatment when they discussed the specific example.

Thank you for this insightful question. While EGFR inhibitors are generally indicated for patients with confirmed EGFR driver mutations, in real-world clinical practice, some patients may receive these treatments without such mutations due to factors like off-label use or clinical judgment. To clarify this, we added a brief statement to the Results.

EGFR inhibitors are primarily used for patients with EGFR mutations, though some in our cohort received them without confirmed mutations due to real-world practices.

Reviewer #4 (Remarks on code availability):

The reviewer checked the github page but did not run the codes.

Reviewer #5 (Remarks to the Author):

Comments to responses

While the authors have adequately addressed the concerns raised by reviewers #2 and #3, I share the concern pointed out by reviewer #2 regarding the novelty of the findings. The current manuscript does not appear to offer significantly new insights when compared to the authors' previous work. The additional predictive mutation-treatment interactions and the machine learning model for risk score prediction, which have limited clinical applicability or validation, may not provide sufficient impact to justify publication in Nature Communications. However, I would defer to the editor's judgment on this matter.

Thank you for the feedback. In this study, we have extended our previous work by analyzing a significantly larger dataset across more cancer types and with longer patient follow-up, enabling us to identify novel mutation-treatment interactions. Additionally, our machine learning model for risk score prediction is presented as a foundation for future clinical validation, contributing new hypothesis-generating insights. We clarified these points in the manuscript to emphasize the unique contributions of this study.

Further comments for consideration

Despite the authors have stated in their response to Reviewer #1 that "The current study did not include an analysis of co-mutations," it would be beneficial to include a simple analysis that could provide valuable insights. A basic visual representation, such as an oncoplot (e.g., https://bioconductor.org/packages/release/bioc/vignettes/maftools/inst/doc/maftools.html#72_Oncoplots), could illustrate co-occurring or mutually exclusivity of predictive mutations across different cancer types. For instance, in a cohort of non-small cell lung carcinoma, patients could be stratified based on the co-occurrence or mutual exclusivity of TP53 and CDKN2A/B mutations. The authors could then demonstrate the differences in the impact on treatment outcomes between these stratified groups.

Thank you for this valuable suggestion. While we have focused our analysis on individual mutation-treatment interactions, we agree that an oncoplot illustrating co-occurring or exclusive mutations could provide useful context. We have added in the Discussion:

Additionally, while this study focuses on individual mutation-treatment interactions, future work could explore co-occurring or exclusive mutations to further contextualize these findings.

Typo

line 135: gene-mutation -> gene-treatment

Thank you. We have updated the text.