

Single-cell and in vivo profiling reveal heterogeneous and organ-specific CRISPR-Cas9 off-target and translocation outcomes

Corresponding Author: Dr Pinar Akcakaya

This manuscript has been previously reviewed at another journal. This document only contains information relating to versions considered at Nature Communications. Mentions of the other journal have been redacted.

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

Version 0:

Reviewer comments:

Reviewer #1

(Remarks to the Author)

The authors have addressed this reviewer's questions and concerns with the revisions to their manuscript.

Reviewer #3

(Remarks to the Author)

The Authors have done a great job in revising their manuscript previously submitted to [Redacted], and have satisfactorily addressed all of my original remarks.

I especially appreciate the Authors's effort in reproducing their analysis on single cells subjected to whole-genome amplification (WGA) and their attempt to perform Nanopore sequencing on some of the WGA samples. (I agree with the Authors, though, that this approach is not scalable as the both the amount of input material required and the cost of Nanopore WGS are still too high).

I also appreciate the Authors' effort in retrieving publicly available Hi-C datasets that could be used to better contextualise the formation of translocations within the 3D genome. However, I understand that it is difficult to find properly matched datasets that could be used to rigorously test the hypothesis that I proposed in my previous review.

In sum, I think the revised manuscript is now suitable for publication in Nature Communications.

Reviewer #4

(Remarks to the Author)

I have been asked to evaluate the Authors' response to the comments originally raised by Reviewer 2.

R2 raised concerns about over-claiming in the first draft (which were echoed by other reviewers) and the authors have been responsive to that. Some of R2's requests for more experimentation, analysis, and speculation struck me as overly-detailed and failed to advance the core argument of the manuscript, so I am less concerned about whether the authors have been fully responsive to every single point. Overall, the experimental approach demonstrated here, now that the results are appropriately caveated and contextualized, will be a nice addition to the literature.

Open Access This Peer Review File is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons license, and indicate if changes were made.

In cases where reviewers are anonymous, credit should be given to 'Anonymous Referee' and the source.

The images or other third party material in this Peer Review File are included in the article's Creative Commons license, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons license and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder.

To view a copy of this license, visit <https://creativecommons.org/licenses/by/4.0/>