



Life Science Alliance

Caspase-1 interdomain linker cleavage is required for pyroptosis

Daniel Ball, Cornelius Taabazuing, Andrew Griswold, Elizabeth Orth, Sahana Rao, Ilana Kotliar, Lauren Vostal, Darren Johnson, and Daniel Bachovchin

DOI: <https://doi.org/10.26508/lsa.202000664>

Corresponding author(s): Daniel Bachovchin, Memorial Sloan Kettering Cancer Center

Review Timeline:

Submission Date:	2020-01-31
Editorial Decision:	2020-02-03
Revision Received:	2020-02-03
Accepted:	2020-02-04

Scientific Editor: Andrea Leibfried

Transaction Report:

(Note: With the exception of the correction of typographical or spelling errors that could be a source of ambiguity, letters and reports are not edited. The original formatting of letters and referee reports may not be reflected in this compilation.)

Please note that the manuscript was previously reviewed at another journal and the reports were taken into account in the decision-making process at Life Science Alliance. Since the original reviews are not subject to Life Science Alliance's transparent review process policy, the reports and author response cannot be published.

February 3, 2020

RE: Life Science Alliance Manuscript #LSA-2020-00664-T

Prof. Daniel A Bachovchin
Memorial Sloan Kettering Cancer Center
Chemical Biology Program
1275 York Avenue
Box 428
New York, New York NY 10065

Dear Dr. Bachovchin,

Thank you for transferring your revised manuscript entitled "Caspase-1 interdomain linker cleavage is required for pyroptosis". Your manuscript was reviewed twice at another journal before, and the editors provided those reviewer reports to us with your permission.

While one reviewer was supportive of publication, two of the reviewers who re-evaluated your study at the other venue would have expected a further reaching conceptual advance and more insight in a physiologically-relevant system. We had indicated to you prior to submission, that such physiologically-relevant insight is not needed for publication here. We would thus be happy to publish your paper in Life Science Alliance pending final minor revisions:

- Please provide a full point-by-point response to the remaining concerns. Please address the remaining concerns of reviewer #1 with text changes. Please address the remaining concerns of reviewer #2 in the following way: if possible, please provide evidence for the GSDMD-p30 band in your blots being indeed p30. Please check carefully throughout your manuscript and add information on replicates. Please mention within the manuscript text or figure legend that cells were not fixed prior to ASC spec analysis and change the quantification to % of cells with ASC specks. Please fix the wording in the abstract to respond to the reviewer concern #4 and address the minor issues.
- Please add a summary blurb and author contributions (to be selected for each author in the author area when submitting) within our submission system
- Please provide all figures, including supplementary figures, as individual files, the legends should remain in the main manuscript file (in a separate section)
- Please provide the main ms file in word docx format

If you are planning a press release on your work, please inform us immediately to allow informing our production team and scheduling a release date.

To upload the final version of your manuscript, please log in to your account:

<https://lsa.msubmit.net/cgi-bin/main.plex>

You will be guided to complete the submission of your revised manuscript and to fill in all necessary information. Please get in touch in case you do not know or remember your login name.

To avoid unnecessary delays in the acceptance and publication of your paper, please read the following information carefully.

A. FINAL FILES:

These items are required for acceptance.

- An editable version of the final text (.DOC or .DOCX) is needed for copyediting (no PDFs).
- High-resolution figure, supplementary figure and video files uploaded as individual files: See our detailed guidelines for preparing your production-ready images, <http://www.life-science-alliance.org/authors>
- Summary blurb (enter in submission system): A short text summarizing in a single sentence the study (max. 200 characters including spaces). This text is used in conjunction with the titles of papers, hence should be informative and complementary to the title. It should describe the context and significance of the findings for a general readership; it should be written in the present tense and refer to the work in the third person. Author names should not be mentioned.

B. MANUSCRIPT ORGANIZATION AND FORMATTING:

Full guidelines are available on our Instructions for Authors page, <http://www.life-science-alliance.org/authors>

We encourage our authors to provide original source data, particularly uncropped/-processed electrophoretic blots and spreadsheets for the main figures of the manuscript. If you would like to add source data, we would welcome one PDF/Excel-file per figure for this information. These files will be linked online as supplementary "Source Data" files.

****Submission of a paper that does not conform to Life Science Alliance guidelines will delay the acceptance of your manuscript.****

****It is Life Science Alliance policy that if requested, original data images must be made available to the editors. Failure to provide original images upon request will result in unavoidable delays in publication. Please ensure that you have access to all original data images prior to final submission.****

****The license to publish form must be signed before your manuscript can be sent to production. A link to the electronic license to publish form will be sent to the corresponding author only. Please take a moment to check your funder requirements.****

****Reviews, decision letters, and point-by-point responses associated with peer-review at Life Science Alliance will be published online, alongside the manuscript. If you do want to opt out of having the reviewer reports and your point-by-point responses displayed, please let us know immediately.****

Thank you for your attention to these final processing requirements.

Thank you for this interesting contribution, we look forward to publishing your paper in Life Science Alliance.

Sincerely,

Andrea Leibfried, PhD
Executive Editor
Life Science Alliance
Meyerhofstr. 1
69117 Heidelberg, Germany
t +49 6221 8891 502
e a.leibfried@life-science-alliance.org
www.life-science-alliance.org

LSA Point-by-point response

While one reviewer was supportive of publication, two of the reviewers who re-evaluated your study at the other venue would have expected a further reaching conceptual advance and more insight in a physiologically-relevant system. We had indicated to you prior to submission, that such physiologically-relevant insight is not needed for publication here. We would thus be happy to publish your paper in Life Science Alliance pending final minor revisions:

- Please provide a full point-by-point response to the remaining concerns.

Our responses are in blue

Please address the remaining concerns of reviewer #1 with text changes.

We have amended the text to include the specific items requested, including the sgRNA used for the CASP1 KO THP-1 cell line and the sequence information of human caspase-1. The identity of the Lentiguide puro vector was already listed in the “cloning” methods section.

Please address the remaining concerns of reviewer #2 in the following way: if possible, please provide evidence for the GSDMD-p30 band in your blots being indeed p30.

The labeled GSDMD-p30 band is indeed p30. VbP is well-characterized to induce the formation of this GSDMD p30 in WT THP-1 cells. These cells were used in this experiment, and this band is clearly visible in the second lane of the gel, which is precisely the correct size. Thus, this is the correct p30 band.

Please check carefully throughout your manuscript and add information on replicates.

We have carefully checked and added information of replicates throughout the manuscript.

Please mention within the manuscript text or figure legend that cells were not fixed prior to ASC spec analysis and change the quantification to % of cells with ASC specks.

We have now noted in the legend that the cells were not fixed prior to speck analysis. Due to the experimental setup, we cannot quantify the % of cells with ASC specks from these images. However, we should note that not all publications quantify % of cells with specks. For example, Boucher et al (JEM, 2018) report the relative intensity of specks per field of view (Fig. 4C, 5A), Van Opdenbosch et al (Nat. Comm. 2014) report #specks/field (Fig. 4B, 4D), and Wickliffe et al (Wickliffe, Nature. 2019) quantitate #specks/region of interest (Fig. 4B). Our analysis is in line with these previous analyses. Moreover, we have corroborated our data with gel-based oligomer assays.

Please fix the wording in the abstract to respond to the reviewer concern #4 and address the minor issues.

We have adjusted this wording.

- Please add a summary blurb and author contributions (to be selected for each author in the author area when submitting) within our submission system

Done

- Please provide all figures, including supplementary figures, as individual files, the legends should remain in the main manuscript file (in a separate section)

[Done](#)

- Please provide the main ms file in word docx format

[Done](#)

February 4, 2020

RE: Life Science Alliance Manuscript #LSA-2020-00664-TR

Prof. Daniel A Bachovchin
Memorial Sloan Kettering Cancer Center
Chemical Biology Program
1275 York Avenue
Box 428
New York, New York NY 10065

Dear Dr. Bachovchin,

Thank you for submitting your Research Article entitled "Caspase-1 interdomain linker cleavage is required for pyroptosis". It is a pleasure to let you know that your manuscript is now accepted for publication in Life Science Alliance. Congratulations on this interesting work.

The final published version of your manuscript will be deposited by us to PubMed Central upon online publication.

Your manuscript will now progress through copyediting and proofing. It is journal policy that authors provide original data upon request.

Reviews, decision letters, and point-by-point responses associated with peer-review at Life Science Alliance will be published online, alongside the manuscript. If you do want to opt out of having the reviewer reports and your point-by-point responses displayed, please let us know immediately.

*****IMPORTANT:** If you will be unreachable at any time, please provide us with the email address of an alternate author. Failure to respond to routine queries may lead to unavoidable delays in publication.***

Scheduling details will be available from our production department. You will receive proofs shortly before the publication date. Only essential corrections can be made at the proof stage so if there are any minor final changes you wish to make to the manuscript, please let the journal office know now.

DISTRIBUTION OF MATERIALS:

Authors are required to distribute freely any materials used in experiments published in Life Science Alliance. Authors are encouraged to deposit materials used in their studies to the appropriate repositories for distribution to researchers.

You can contact the journal office with any questions, contact@life-science-alliance.org

Again, congratulations on a very nice paper. I hope you found the review process to be constructive and are pleased with how the manuscript was handled editorially. We look forward to future exciting submissions from your lab.

Sincerely,

Andrea Leibfried, PhD
Executive Editor
Life Science Alliance
Meyerhofstr. 1
69117 Heidelberg, Germany
t +49 6221 8891 502
e a.leibfried@life-science-alliance.org
www.life-science-alliance.org