

Peer Review Information

Journal: Nature Genetics

Manuscript Title: Alternative splicing during mammalian organ development

Corresponding author name(s): Professor Henrik Kaessmann

Editorial Notes:

Transferred manuscripts This manuscript has been previously reviewed at another journal that is not operating a transparent peer review scheme. This document only contains reviewer comments, rebuttal and decision letters for versions considered at Nature Genetics.

Reviewer Comments & Decisions:

Decision Letter, initial version:
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Our ref: NG-AN55775-T

2nd Feb 2021

Dear Dr. Kaessmann,

Thank you for submitting your revised manuscript "Alternative splicing during mammalian organ development" (NG-AN55775-T). It has now been seen by the original referees and their comments are below. The reviewers find that the paper has improved in revision, and therefore we'll be happy in principle to publish it in Nature Genetics, pending minor revisions to comply with our editorial and formatting guidelines.

***Please note-

In order to proceed, we ask that you please send a word.doc version of the manuscript text, methods text, figure legend text and references. No figures or extended data figures need to be included. This will aid us in our formatting checks.

Can you please send this to me as soon as possible?

We are now performing detailed checks on your paper and will send you a checklist detailing our editorial and formatting requirements in about a week. Please do not upload the final materials and

make any revisions until you receive this additional information from us.

Thank you again for your interest in Nature Genetics Please do not hesitate to contact me if you have any questions.

Congratulations on the paper!

All the best,

Catherine

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Reviewer #1 (Remarks to the Author):

Note : This is my second revision of this manuscript (previously Referee #3).

The manuscript entitled "Alternative splicing during mammalian organ development", by Mazin et al., presents a comparative analysis alternative splicing during development (devAS) in 7 species (6 mammals and chicken). The analysis is thorough and comprehensive, ranging from AS developmental patterns and their conservation, the apparition and evolution of new AS events, and associated genome structures (microexons, DNA motifs and transposable elements). It is the first AS analysis encompassing such a large dataset in development. The manuscript is clearly written. I think the manuscript is a very valuable contribution to the open debate on the functional importance of AS in mammalian genomes. This manuscript represents a huge work, the data is carefully analyzed and is a very useful resource for future work.

In my first revision, I only had a couple of requests, mainly to clarify and quantify some verbal assessment in the manuscript. The authors have answered these completely, in a very thorough and complete manner, and sometimes beyond my expectancies. I would like to thank them for this effort.

A number of figures and complementary analyses have been made, in order to answer the remarks of the 3 referees. I think the addition of the Extended Data Fig1 is very efficient to show the difference of developmental sampling across species, and was a very good idea. The authors also assessed, by subsampling analyses, how differences in developmental sampling might explain differences in the amount of detected AS and devAS events between species. They show convincingly that this is not the case (new Extended Data Fig. 4 & 14).

Regarding the question of tissue composition : I appreciate that it is not possible to deconvolve the

devAS patterns of the testis developmental bulk-tissue using cell type-specific RNA-seq data. The authors made it clear in their rebuttal and in the new text. They also acknowledge in the text, that "one important limitation in that the underlying bulk-tissue RNA-seq data do not generally allow us to assess the relative contributions of changes in cellular composition versus change".

I'm also fully convinced by the answers to my other questions, and to the answer to the questions from the 2 other reviewers.

Reviewer #2 (Remarks to the Author):

The authors satisfactorily addressed most of my previous comments. It is acceptable for publication on Nature Genetics in my opinion and this work will be a great resource for the community.

Final Decision Letter:

In reply please quote: NG-AN55775R Kaessmann

19th Mar 2021

Dear Dr. Kaessmann,

I am delighted to say that your manuscript "Alternative splicing during mammalian organ development" has been accepted for publication in an upcoming issue of Nature Genetics.

Prior to setting your manuscript, we may make minor changes to enhance the lucidity of the text and with reference to our house style. We therefore ask that you examine the proofs most carefully to ensure that we have not inadvertently altered the sense of your text in any way.

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Congratulations on the paper!

All the best,

Catherine

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