**Reviewer’s report**

**Title:** Blocking podoplanin suppresses growth and pulmonary metastasis of human malignant melanoma

**Version:** 1  **Date:** 19 Feb 2019

**Reviewer:** Teresa Cardoso

**Reviewer’s report:**

The authors describe how podoplanin (PDPN) is involved in tumor growth and metastasis. They also evaluated the effect of a functional blocking monoclonal antibody (SZ168) that has been previously developed by the same authors. Even though the set of experiments shown are well-written and presented the major drawback of the study is the fact that it is difficult to assess in vivo the metastatic potential of a primary tumor showing different growth rates. This limitation of the study should be at least discussed.

In Figure 1. From the Western blot depicted in Fig 1 C and D there are two bands (25 and 36 KDa) that correspond to PDPN. Can the authors elaborate on this. Are there two isoforms of the protein?

Other question is relative to the fact that the antibody SZ163 is not able to recognize the 25KDA PDPN band. Why is that? Could this be somehow related to the fact that this antibody is not able to reduce the platelet aggregation rate?

Finally, the authors should explain why they have used EPR7072 as a control as it only recognizes the 25KDa band.

In Figure 4A it should be better depicted that the second panel is a zoom of the tumors or metastasis shown in the first panel. Also, it should be at least stated form which animals are this primary tumors shown. I believe that form the CHO/hPDPN + Mouse IgG.

Fig. 7A. It should be better shown or explained that the second panel is a zoom of the metastasis shown in the first panel.

Typos: in the Methods section of the abstract it should be read "… inoculating subcutaneously human malignant…"

**Are the methods appropriate and well described?**
If not, please specify what is required in your comments to the authors.

Yes

**Does the work include the necessary controls?**
If not, please specify which controls are required in your comments to the authors.
No

Are the conclusions drawn adequately supported by the data shown?
If not, please explain in your comments to the authors.

No

Are you able to assess any statistics in the manuscript or would you recommend an additional statistical review?
If an additional statistical review is recommended, please specify what aspects require further assessment in your comments to the editors.

I am able to assess the statistics

Quality of written English
Please indicate the quality of language in the manuscript:

Acceptable

Declaration of competing interests
Please complete a declaration of competing interests, considering the following questions:

1. Have you in the past five years received reimbursements, fees, funding, or salary from an organisation that may in any way gain or lose financially from the publication of this manuscript, either now or in the future?

2. Do you hold any stocks or shares in an organisation that may in any way gain or lose financially from the publication of this manuscript, either now or in the future?

3. Do you hold or are you currently applying for any patents relating to the content of the manuscript?

4. Have you received reimbursements, fees, funding, or salary from an organization that holds or has applied for patents relating to the content of the manuscript?

5. Do you have any other financial competing interests?

6. Do you have any non-financial competing interests in relation to this paper?

If you can answer no to all of the above, write 'I declare that I have no competing interests' below. If your reply is yes to any, please give details below.

I declare that I have no competing interests.

I agree to the open peer review policy of the journal. I understand that my name will be included on my report to the authors and, if the manuscript is accepted for publication, my named report
including any attachments I upload will be posted on the website along with the authors' responses. I agree for my report to be made available under an Open Access Creative Commons CC-BY license (http://creativecommons.org/licenses/by/4.0/). I understand that any comments which I do not wish to be included in my named report can be included as confidential comments to the editors, which will not be published.

I agree to the open peer review policy of the journal