World Journal of Stem Cells

Manuscript 47333 – Steichen et al.

Response to reviewers

Reviewer 1: ID 02937551

Conclusion: Accept (High priority)

Scientific Quality: Grade B (Very good)

Language Quality: Grade A (Priority publishing)

The paper by Steichen et al describes the type of genomic abnormalities observed in hiPSCs, the impact of reprogramming parameters and differentiation protocols on the maintenance of the cellular genomic integrity, and the impact of genomic alterations on the possible usages of hiPSCs and their derivatives. The review is strongly recommended for publication.

We sincerely thank the reviewer for these very positive comments.

Reviewer 2: ID 00546602

Conclusion: Accept (General priority)

Scientific Quality: Grade B (Very good)

Language Quality: Grade B (Minor language polishing)

This manuscript, 'The genomic integrity of human induced pluripotent stem cells (hiPSCs): from reprogramming to differentiation and its subsequent impact for hiPSC-based cell therapy' describes in a concise manner, the problems arising with iPSCs being prone to genetic instability and being linked to tumorigenicity, and discusses the quality control methods implemented to enforce hiPSC genomic integrity. It summarises aptly the latest developments in the field covering all aspects that were not very clear in literature before, particularly comparing data with human embryonic stem cells used as gold standard. The literature is critically analysed and is up-to-date and inferences drawn are reasonable. Highlights from this review will go a long way to assist in optimising protocols and developing therapy with these cells down the track. Strong point: The genomic integrity quality control for hiPSCs and complexities associated with predicting the subsequent impact of genomic abnormalities are very well written and covers all aspects

We sincerely thank the reviewer for these very positive comments.

In terms of their own liver therapy experiment, the authors have not clearly outlined the reason behind no CNV being triggered using their in-house differentiation protocol. The review would be stronger if the authors a) Elaborate why and how no de novo CNV were triggered? b) Account for CNVs acquired during natural selection . I have no hesitation in recommending this review for publication with minor changes

We thank the reviewer for this comment.

Indeed, the natural selection reveals "acquisition" of CNVs because the reprogramming occurred in one precise cell which, for n reasons during the life of the donor, had acquire a CNV, which is thus propagated to the progeny without any selection AND/OR because a CNV occurred during the reprogramming process (at an early phase) and had been selected during the almost 100 mitosis and 3 months necessary to reach passage 10 because of a survival/proliferative advantage. The selection, linked to the number of mitosis and the time in culture, allowed a sufficient take-over of the cells bearing the CNV making its detection possible.

In our hepatic differentiation protocols, in contrast, no more than 15 mitosis and 20 days in culture distinguish between undifferentiated and differentiated cells, which is probably no enough to allow emergence of a detectable clone of cells that could have acquire a new CNV except if a huge selective advantage exist but the latter was not observed (at that time, and no more since, in our experience).

In order to fulfill reviewer's requirement, we added one sentence in the manuscript (page 19, highlighted in red in the revised version):

Lastly, in the context of liver therapy, using three different hepatic differentiation experiments, we demonstrated that no de novo CNV were triggered using our differentiation protocol (Steichen et al., 2014) but the time scale (22-24 days) of our differentiation protocol probably does not allow emergence of detectable CNV due to the limited number of mitosis.

We do think that this additional information was worthwhile to add and therefore thank the reviewer for this comment.