January 8, 2021

Professor Lian-Sheng Ma, Science Editor, Company Editor-in-Chief

Dear Prof. Ma,

Thank you very much for your email communication on December 25, 2020 regarding our manuscript “Mesenchymal stromal cell-dependent immunoregulation in chemically induced acute liver failure” (Manuscript No.: 59909, Review). We are grateful for both you and the editors` very pertinent and constructive comments and suggestions, and would like to specifically address the points raised by them as follows:

Reviewers' Comments:

Reviewer #1:

Specific Comments to Authors:

Zhou and colleagues have elegantly reviewed an enigmatic but important topic of mesenchymal stromal cells dependent immune regulation in drug induced liver failure by summarizing animal models. The Table and Figures are very well made and with updated current literature. The manuscript is well written and require minor language and grammar/syntax corrections.

Response: We thank the reviewer for the valuable comments on our work. We have revised the grammar, layout, etc. of the manuscript and all the changes to the content are indicated in red in the revised manuscript.

I suggest, if possible, and where available, the following data: 1. MSCs use in humans with acute liver failure - provide a description and summary para on this, if data is available. 2. Provide a paragraph on current lacunae and future prospects in bench to bedside research on MSC use in ALF and obstacles faced and possible solutions to these.

Response:

1. We appreciate the reviewer’s suggestions. We have searched the databases for the clinical trials of MSCs used in drug induced-acute liver failure. However, there

is little research on this aspect. There are only some recruiting programs of “Mesenchymal Stem Cell Transplantation for Acute-on-chronic Liver Failure”, and the main cause is hepatitis B virus infection, which is different from the subject we reviewed.

Our retrieval process includes:

1) PubMed (Medline)

Search: ("Mesenchymal Stem Cells"[Mesh]) AND "Liver Failure, Acute/chemically induced" [Mesh]

Filters: clinical trails

2) Cochrane Library

Search:

#1 MeSH descriptor: [Mesenchymal Stem Cells] explode all trees

#2 MeSH descriptor: [Liver Failure, Acute] explode all trees with qualifier(s) chemically induced

#3 #1 and #2

3) EMBASE

#1 'mesenchymal stem cell'/exp AND 'acute liver failure'/exp

#2 #1 AND 'human'/de

#3 #2 AND 'clinical trial'/de

4) SinoMed CBM

("Mesenchymal Stem Cells" AND "acute liver injury") AND ("clinical trails"[literature type])

5) ICH GCP (http://ichgcp.net/clinical-trials-registry/)

"Mesenchymal Stem Cells" AND "acute liver injury"

2. Following the Reviewer’s comments, we have added a paragraph on current lacunae and future prospects in “Conclusions and Future Directions” section. Changes to the content are indicated in red in the revised manuscript and we also show them here as follows:

However, there are still some deficiencies in the research of MSC-dependent immunoregulation in chemically induced ALF. For example, the pathogenesis of liver-injury models and the role of the immune system are still unclear. There has not been enough extensive and in-depth research on MSC-dependent immunoregulation in chemically induced ALF.

Different sources and different pretreated MSCs have varying therapeutic effects on liver injury. To date, there is no uniform standard for MSC applications in animal models. Thus, the results from different studies cannot be compared or repeated in different laboratories under different conditions.

MSCs have been widely studied for their differentiation and immunomodulation abilities. However, in one study, researchers focused on a single capability of MSCs, ignoring comparisons of their various capabilities. Future studies are needed to determine which MSC capability dominates.

There have been no clinical trials on DILI treated by MSCs. Clinical trials on MSC treatment are often applied to chronic diseases such as GVHD, diabetes, and malignant blood disease. MSCs are rarely used in DILI, which has a rapid onset and high mortality rate, and more conventional and conservative treatments tend to be used. Clinical trials can be conducted only if the efficacy and safety of MSCs are supported by sufficient research. The two main obstacles to translating the results from animal experiments into clinical practice are that the pathogenicity of ALI caused by clinical drugs differs from that of animal models, and that the immune system of animals, such as mice, is different from that of humans, so the results demonstrated in mice are not necessarily applicable to humans. Possible solutions to these issues are to verify the results obtained in animal experiments in organoids derived from human liver, and to identify animal models with a similar pathogenicity to DILI in humans. Further studies are needed to reveal the therapeutic mechanisms of MSCs.

Taken together, we feel that your comments and suggestions are very helpful in improving our manuscript, and therefore have made corresponding additions and modifications (marked in red in the re-submitted version). Please let us know if you

need further clarification. Thank you once again for your professional and timely assistance.

Sincerely,

Hongcui Cao