

Dear Editors and Reviewers:

Thank you for your letter and for the reviewers' comments concerning our manuscript entitled "MKK7-mediated phosphorylation of JNKs regulates the proliferation and apoptosis of human spermatogonial stem cells" (69911). Those comments are all valuable and very helpful for revising and improving our paper, as well as the important guiding significance to our researches. We have studied comments carefully and have made correction which we hope meet with approval. Revised portion are marked in red in the paper. The main corrections in the paper and the responds to the reviewer's comments are as flowing:

Responds to the reviewer's comments:

Reviewer #1:

1. Response to comment: Material and Methods: Please clearly and specifically describe the method or procedure of specimen collection and sample size for each group in detail. How many patients involved, donor selection criteria, method of specimen collection, the portion of specimen collected (Is it a whole specimen, both, one part, partial, or a biopsy size?). Was the specimen taken from deceased patients or living ones? please clarify.

Response: We are very sorry for this negligence, all of testis samples were derived from patients undergoing microdissection testicular sperm extraction, and patients with spermatogenic failure because of known hereditary factors, such as Klinefelter syndrome and Y chromosome microdeletions, were excluded. We collected a total of 16 testicular biopsies weighing 30-50 mg and classified them according to the results of HE staining, including normal, spermatogenic failure, and Sertoli cell only syndrome. Revised portion are marked in red in the paper.

Line 111-117: The statements of "All of testis samples were derived from patients undergoing microdissection testicular sperm extraction, and patients with spermatogenic failure because of known hereditary factors, such as Klinefelter syndrome and Y chromosome microdeletions, were excluded. We collected a total of

16 testicular biopsies weighing 30-50 mg and classified them according to the results of HE staining, including normal, spermatogenic failure, and Sertoli cell only syndrome.” were added.

2. Response to comment: Please provide the informed consent from the donors or patients. How do the patients provide their tissue samples?

Response: Considering the Reviewer’s suggestion, We have provided the ethical review approval and informed consent of some patients in the attachment, and we are happy to undergo ethical review at any time. We collected the remaining testicular tissue after microdissection testicular sperm extraction without affecting the patient's treatment and obtained the patient's consent.

3. Response to comment: Regarding the ethics, please provide strong argument on the priority and necessity of your study.

Response: We always adhere to the principle of ensuring the treatment priority of patients and collect specimens remaining after microdissection testicular sperm extraction for scientific research. Our research on human spermatogonial stem cells will certainly be an important direction for the recovery and treatment of male fertility in the future, and also provide a target for the diagnosis of male infertility and the development of contraceptives.

4. Response to comment: Please check and correct the mistyped words.

Response: Thanks to the reviewers for their suggestions, we revised the spelling errors in the text and marked the revised parts in red.

5. Response to comment: Please check and use the SI units.

Response: We are very sorry for this negligence, we made modification to all the unit labeling according to the requirements of SI units, and the modified part is marked in red.

6. Response to comment: Statistical Analysis: please provide a formal certificate of

biostatistics.

Response: Considering the Reviewer's suggestion, we provided a formal certificate of biostatistics in the attachment.

7. Response to comment: Results: Please explain more comprehensively the result of HPA analysis among the groups (Figure 6A).

Response: Thanks to the reviewers for their suggestions, we described Figure 6A in more detail, and the modified part was marked in red.

Line 341-line 344: The statements of "According to the results of hematoxylin and eosin (HE) staining, we confirmed the spermatogenesis status of the testis (Fig S2A-S2H). We examined the positive proportion and localization changes of MKK7 (red) in SSCs using double immunohistochemistry with UCHL1 (green)" were added.

Line 348: The statements of "but there were no translocations of MKK7 protein were observed in NOA samples." were added.

8. Response to comment: References: Please arrange the reference list according to the guidelines for authors

Response: We are very sorry for this negligence, we modified the format of the references according to the requirements of the authors' guidelines.

Additional revisions to the manuscript include:

With the consent of all authors, we added one affiliation and grant funding.

Line 13 and 14: The statements of "c. Clinical Research Center for Reproduction and Genetics in Hunan Province, Changsha, Hunan, People's Republic of China." were added.

Line 432 and 433: The statements of "Hunan Provincial Grant for Innovative Province Construction (2019SK4012)." were added.

Special thanks to you for your good comments!

We tried our best to improve the manuscript and made some changes in the manuscript. These changes will not influence the content and framework of the paper. And here we did not list the changes but marked in red in revised paper.

We appreciate for Editors/Reviewers' warm work earnestly, and hope that the correction will meet with approval.

Once again, thank you very much for your comments and suggestions.