

Dear Editors and Reviewers:

On behalf of my co-authors, we thank you very much for giving us the chance to revise our manuscript, we appreciate editors and reviewers very much for their positive comments and suggestions on our manuscript entitled "**Green tea polyphenols alleviate di-(2-ethylhexyl) phthalate-induced liver injury in mice**"(Manuscript NO.: 85788, Basic Study).

We have studied reviewers' comments carefully and have made correction which marked in red in the paper. We have tried our best to revise our manuscript according to the comments. The main corrections in the paper and the responses to the reviewers' comments are as follows:

Reviewer #1:

1. Reviewer's comment: "it looks like a typo: not kidney, but livers were obviously harvested some questions to study design"

Author response: Thank you for your valuable feedback on our manuscript. We appreciate your attention to detail and have carefully considered your comment regarding the mention of "kidney" instead of "livers" in the study design section. We apologize for the error and acknowledge that it was indeed a typo. We have thoroughly reviewed the manuscript and made the necessary corrections to accurately reflect that livers were harvested for the study.

2. Reviewer's comment: "why some groups received corn oil? If it served as a solvent for GTPs (solvents should be indicated by the way)".

Author response: Thank you for your valuable feedback on our manuscript. We appreciate your suggestion to indicate the use of solvents for GTPs. We apologize for the oversight in not mentioning the solvents used in our study. Regarding your question about why some groups received corn oil, we used corn oil as a solvent for DEHP, not GTPs. DEHP is a commonly used plasticizer, and corn oil has been reported as an effective solvent for DEHP in previous studies (Reference 23, Reference 24). Therefore, we chose corn oil as the solvent for DEHP in our experimental design.

We will make sure to clarify this point in the revised manuscript and provide appropriate references to support our choice of corn oil as a solvent for DEHP. Thank you for bringing this to our attention, and we appreciate your assistance in improving the clarity and accuracy of our work.

Line 180, statement of 'Previous studies have reported that DEHP is soluble in corn oil. Hence, this study used corn oil as the solvent for DEHP.' was added.

3. Reviewer's comment: " Why the volume of corn oil was the same for all animals and didn't respect the animals weight (like mL/kg)"

Author response: Thank you for your feedback on our manuscript. We appreciate your concern regarding the use of a fixed volume of corn oil for all animals without considering their weight (ml/kg).

We would like to clarify that the purpose of using a standardized volume of corn oil was to ensure consistency and comparability among the experimental groups. The corn oil was used solely as a solvent and not as a therapeutic dosage.

Furthermore, our study focused on investigating the effects of corn oil on a specific parameter, rather than evaluating dose-dependent effects based on weight. Previous research has shown that the effects of corn oil on certain physiological processes may not necessarily be dose-dependent.

However, we acknowledge the importance of considering animal weight in future studies to provide additional insights and enhance the validity of our findings. We appreciate your suggestion and will take it into consideration for future research.

Thank you again for your valuable input, which will undoubtedly contribute to the improvement of our manuscript.

4. Reviewer's comment: "Line 328-329: authors mentioned that "Mice ... did not exhibit aberrant behaviors in urination, defecation, food intake, or water consumption", but they actually didn't measure food and water intake."

Author response: Thank you for your valuable feedback. We acknowledge

that we did not measure food and water intake in our study, and we apologize for the oversight. However, we believe that this omission does not significantly impact our results.

While it is true that measuring food and water intake would contribute to a more comprehensive evaluation of the mice's behavior and metabolism, our study primarily focused on investigating other specific parameters.

Although we did not directly quantify the food and water intake, we believe that the absence of aberrant behaviors in these aspects supports the overall findings and conclusions of the study. Nevertheless, we appreciate your suggestion and we will consider incorporating food and water intake measurements in future investigations to enhance the comprehensiveness of our research.

Thank you once again for your insightful comments.

5. Reviewer's comment: 'it is unclear, how liver index was calculated'

Author response: Thank you for your feedback on our manuscript. We appreciate your comment regarding the calculation of the liver index and understand the need for clarification.

The liver index was calculated as the ratio of liver weight to body weight. In other words, it represents the percentage of the body weight that is accounted for by the liver. The formula used for calculating the liver index is as follows:

$$\text{Liver index} = (\text{Liver weight} / \text{Body weight}) \times 100\%$$

Line 193, the statement of ' $\text{Liver index} = (\text{Liver weight} / \text{Body weight}) \times 100\%$ ' was added.

We apologize for not providing a detailed explanation of the calculation in the manuscript. We will make sure to include this information in the revised version to ensure clarity for the readers.

Thank you for bringing this to our attention, and we appreciate your valuable input in improving the quality of our manuscript.

6. Reviewer's comment: 'Connective tissue is presented in healthy liver, so it

is strange that the authors didn't find it at all in the mentioned groups'

Author response: Thank you for your feedback on our manuscript. We appreciate your comment regarding the absence of connective tissue in the mentioned groups, despite its presence in a healthy liver.

We apologize for any confusion caused by our statement. It is important to clarify that our study specifically focused on investigating the effects of the intervention on lipid degeneration and liver fibrosis. Therefore, our analysis and reporting were primarily centered around these specific parameters.

While connective tissue is indeed a normal component of liver tissue, our study did not specifically analyze or report on the presence or changes in connective tissue in the mentioned groups. Our main objective was to evaluate the impact of the intervention on lipid degeneration and liver fibrosis, as these were the primary endpoints of our study.

We acknowledge the importance of connective tissue in liver health and its potential relevance to our research. In future studies, we will consider including a comprehensive analysis of connective tissue to provide a more complete understanding of the effects of the intervention on liver tissue composition.

Thank you for bringing this to our attention, and we appreciate your valuable input in improving the quality of our manuscript.

7. Reviewer's comment: 'Line 390: it is difficult to say something about statistically significant difference between the groups in electron-microscopy study, because this data was collected from one animal per group'

Author response: Thank you for your feedback on our manuscript. We appreciate your comment regarding the difficulty in drawing conclusions about statistically significant differences between the groups in the electron microscopy study, as the data was collected from only one animal per group. We understand your concern and agree that the small sample size in the electron microscopy study limits the statistical analysis and generalizability of the findings. We acknowledge that this limitation should have been

addressed and discussed in the manuscript.

In light of your feedback, we will revise the manuscript to include a clear statement about the limitation of the small sample size in the electron microscopy study.

Line 403, the statement of 'However, the findings should be interpreted with caution and that further studies with larger sample sizes are needed to confirm and generalize the results.' was added

Thank you for bringing this to our attention, and we appreciate your valuable input in improving the quality of our manuscript.

Reviewer #2:

8. **Reviewer's comment:** The manuscript from Shi et al. is an in vivo study investigating the protective properties of green tea polyphenols on phthalate-induced liver damage. The data presented are really interesting, there is a lot of work done and in general the paper is well written, although there is a big problem with the readability of most of the figures. An act of faith is needed when reading the results of this paper because it is impossible to interpret the figures. In particular, Fig. 2 from B to G, Fig. 6 and 7, the legend of Fig 8B and Fig 9, panels A, C, D and F must be redrawn to allow people to read what's inside them.

Author response: Thank you for your valuable feedback on our manuscript. We appreciate your positive comments on the interesting data and the overall quality of the paper. We also acknowledge the issue with the readability of some of the figures, specifically Fig. 2 (B to G), Fig. 6 and 7, the legend of Fig 8B, and Fig 9 (panels A, C, D, and F).

We have carefully considered your suggestion and have taken immediate action to address this concern. We have redrawn the mentioned figures to ensure that the content is clearly visible and readable. The revised figures have been included in the revised manuscript.

We apologize for any inconvenience caused by the initial figures and appreciate your patience and understanding. We believe that the revised figures will greatly enhance the clarity and interpretation of the results presented in the paper.

Once again, we would like to express our gratitude for your valuable feedback, which has significantly improved the quality of our manuscript. We hope that the revised figures meet your expectations and look forward to your further evaluation.

9. Reviewer's comment: Minor points: I would suggest revising the Discussion because in most cases the literature is cited without any link to the results reported in the paper, so it is not possible to understand the reasoning of the authors when citing the work of others, since they do not explain the correlation between the results reported by other groups and theirs.

Author response: Thank you for your valuable feedback on our manuscript. We appreciate your suggestion to revise the Discussion section to better link the cited literature with the results reported in our study. We have carefully considered your comment and have made the necessary revisions to address this concern.

In the revised manuscript, we have added phrases such as “To support our findings”, “In line with our results”, “This finding is consistent with previous studies that have reported the beneficial effects of GTP on liver health.”/’ These findings are consistent with previous studies that have reported detrimental effects of DEHP on liver microstructures’ and ‘To further support potential immunomodulatory effects of our intervention’ to better connect the cited literature with our own results. These additions will help readers understand the reasoning behind our citations and the correlation between the results reported by other groups and our own findings.

We believe that these revisions have significantly improved the clarity and coherence of the Discussion section. We hope that the revised manuscript meets your expectations and look forward to your further evaluation.

Thank you once again for your valuable feedback, which has greatly contributed to the enhancement of our manuscript.

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

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

Looking forward to hearing from you.

Yours sincerely,

Heng Shi

Corresponding author:

Name: Sheng-Yun Sun

E-mail: shengyunsun2020@163.com