# ANSWERING REVIEWERS

## *Aug 4, 2022*

## Dear Editors and Reviewers,

Please find enclosed the edited manuscript in Word format (**WJG-NO-78313**).
Title: **Mitochondrial CPT-II Dysfunction: A Possible Novel Mechanism for NAFLD in Hepatocarcinogenesis**

Author: **Min Yao, Ping Zhou, Yan-Yan Qin, Li Wang and Deng-Fu Yao**

Name of Journal: **World Journal of Gastroenterology**

## Reviewer 1 comments:

**Thanks to you for your very kindly comments.**

The authors present a review of the progress of CPT-II inactivity with liver fat accumulation as possible novel pathogenesis for NAFLD in hepatocarcinogenesis in the manuscript “Mito- chondrial CPT-II Dysfunction: A Possible Novel Mechanism for NAFLD in Hepato- carcinogenesis”. The authors make a strong case that CPT-II inactivity has an important role in the NAFLD malignant transformation. The wording and sentence structure also needs work, but overall, it is an interesting paper.

**Major points:**

1. CPT1 has been recognized as the rate-limiting enzyme of the FAO pathway in the past, and the research on CPT2 is the rate-limiting enzyme of this pathway ware relatively few. The author should increase documents to corroborate the view that CPT2 is the rate-limiting enzyme of the FAO pathway.

**Yes, some related-papers has been added.**

① Han S, et al. CPT1A/2-Mediated FAO Enhancement-A Metabolic Target in Radio-resistant Breast Cancer. Front Oncol. 2019; 9:1201. DOI: 10.3389/fonc. 2019.01201. Authors found that CPT1 and CPT2, a pair of rate-limiting enzymes for mitochondrial fatty acid transportation, play a critical role in increasing fatty acid oxidation

② Park JH, et al. Fatty Acid Oxidation-Driven Src Links Mitochondrial Energy Reprogramming and Oncogenic Properties in Triple-Negative Breast Cancer. Cell Rep. 2016; 14(9): 2154-2165. DOI: 10.1016/j.celrep.2016.02.004. Mani- pulation of FAO including the knocking down of CPT1 and CPT2, the rate-limiting proteins of FAO, and analysis of patient-derived xenograft models confirmed the role of mitochondrial FAO in Src activation and metastasis.

2. The first sentence of the second paragraph of the article "1. CPT2 structure" should be pointed out to "facilitates the transfer of what". In previous studies (For example: Current issues regulating treatment of mitochondrial fatty acid oxidation disorders), bezafibrate can be used to treat long-chain FAODs diseases and improve the symptoms of CPT2 deficient diseases. The conclusions inferred in “reference 30” are not particularly consistent with the examples. The author should further enrich the related evidence and merge this paragraph with the “3.1 CPT2 mutation”.

**Thank you very much. This is a nice suggestion, we have further enrich the evidences and merge this paragraph with“the part of CPT2 mutation.**

3. In “2. Mitochondrial CPT system”, the theme of "CPT system" should be highlighted, and the content of this part should be further simplified. Moreover, the term "CPT system" is relatively rare, which is generally called "carnitine shuttle system", the author should consider which model is more appropriate.

**OK. Indeed, the carnitine shuttle system is widely used and the use of the CPT system is not standardized, and this article adopts the carnitine shuttle system, thank you.**

4. It should be pointed out which "fatal human diseases" are included and whether they are related to HCC induced by NAFLD in the last sentence of the second paragraph of 3.3 (Mutation or dysregulation of CPT-II has been linked to many serious, even fatal human diseases, and these data are promising targets for the development of therapeutic agents against NAFLD in future).

**Those fatal human diseases illustrates the importance of FAO during fasting and in hepatic and (cardio) muscular function that has been inserted the related parts. Clinical presentation of CPT-II mutation or dysregulation may include the reported muscle, severe infantile, lethal neonatal forms, hypoketotic hypoglycemia, (cardio) myopathy, arrhythmia, rhabdomyolysis, fatal or handicapped virus-associated encephalopathy and so on, these data are promising targets for the development of therapeutic agents against NAFLD in future.**

5. In the part of “3.4 carnitine level”, the level of carnitine is indeed a known key factor affecting the metabolic flux of the FAO pathway, and the decrease of carnitine concentration observed in NAFLD does not directly indicate that CPT2 is related to the pathogenesis of NAFLD. The effect of carnitine on other FAO enzymes or other pathways in the body is worth explaining or pointing out.

**Yes, you are right. Circulating carnitine level in NAFLD patients was lower than**

**those in healthy people, and the level in NAFLD cases with liver cirrhosis accounted for only 22 %. if patients with HBV or HCV infection, or mitochondrial FAODs present with NAFLD or severe liver diseases, carnitine in the carnitine shuttle system should play an important role, suggesting that enough carnitine levels affect β-oxidation, improves mitochondrial dysfunction, and reduces insulin resistance to ameliorate NAFLD progression.**

6. In “Conclusions and perspectives”, it may not be suitable to describe that "… suggesting that CPT-II might become a new mechanism of blocked lipid oxidation for HCC.", because the expression level or enzyme activity of CPT-II is difficult to analyze, which CPT-II is located in the mitochondrial inner membrane of hepatocytes. Is it appropriate to take CPT-II as an early monitoring index of NAFLD malignant transformation? The author should consider a more reasonable expression, or point out the limitations of CPT-II as an early monitoring index.

**Thank you. This part has been changed and inserted some relevant and meaningful content has been inserted.**

**Minor points:**

Typos and grammatical errors should be corrected, and some expression could be modified. For examples, the expression of singular and plural in the paper; the sentence contains a series of three or more words, phrases or clauses, please insert a comma to separate the elements; “dysfuction” in “Abstract” should be “dysfunction”; “progression” in “Introduction” should be “progress”; the sentence that “Lipid metabolism rearrangements in NAFLD contribute to disease progression that has emerged as one of the most risk for HCC” in “Introduction” should be “Lipid metabolism rearrangements in NAFLD contribute to disease progression that has emerged as one of the most risks for HCC”.

**Those have been corrected, thanks to you.**

# Reviewer 2 comments:

**Thanks to you for your very kindly comments.**

A proactive review of pathophysiology which may be linked to treatment modalities of NAFLD. Please correct typographic errors such as: dysfuction (dysfunction), acyltransferases (acyltransferases), malony-CoA(malonyl-CoA), mutanted (mutated), encephalo pathy (encephalopathy), diseaseallele (disease allele), senother-apeutics

**Thank you very much. Recheck again and these has been corrected.**

We also appreciate the reviewers’ careful and thoughtful suggestions, since the comments are all valuable and helpful for improving our paper. We have studied

comments and made some modifications according to the reviewers' comments or suggestions

Thank you again for publishing our manuscript in the **WJG**.

Sincerely yours,

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