

Response to reviewer for manuscript #60049

Reviewer 1: The authors tried evaluate the anti-NAFLD effect of papaya in high fat diet induced obesity in rats, and concluded that papaya can suppress the lipogenic pathway, improve the balance of antioxidant status and lower systemic inflammation. The study is well designed.

Comments: #1. The authors need to review the literature and discuss any health benefits in general population and individuals with obese and other health issues. Any clinical and human population data to support your study. E. g. Is there any difference in papaya-producing countries/regions compared to people in countries/regions without papaya?

Response: We appreciate your suggestion and have added more information regarding papaya health benefits to the revised MS.

From your point – is there any difference in papaya-producing countries/regions compared to people in countries/regions without papaya? We've looked through the review articles for the association of obesity/NAFLD and papaya. We have found no reports for this relationship. The United States and Europe have increased their importation of papaya during the past decades, especially Germany, Netherlands, Spain, Portugal, and United Kingdom. Their large suppliers are from South America and Asia which are the major cultivation areas (Tridge, 2020). However, the prevalence of NAFLD and obesity are increasing each year and have been found in all parts of the world including the United States, Asia, Middle East and Europe (Fan, et al., 2017; Mitra, et al., 2020; Younossi, 2019). Therefore, there is no scientific evidence suggesting the association between obesity, NAFLD and papaya consumption. Even in the lack of this association, there are a numbers of studies showing the medicinal properties of papaya including the treatment of gastrointestinal related disorders, diabetes, hypertension, hypercholesterolemia, hepatotoxicity, and possess the anti-microbial, anti-parasitic and anti-viral properties (Aravind G, et al., 2013; O'Hare and Williams, 2014; Santana, et al., 2019). Almost all parts of papaya can be used especially the pulp that contain potential antioxidant vitamins – vitamin A, C and E, and other bioactive compounds. However, few studies have been conducted in experimental models specific to obesity. Our present study showed the evidence in an experimental model to demonstrate the therapeutic potential of papaya in NAFLD. Since papaya is low cost and is easily available, widely marketed worldwide, therefore, this can contribute to the implement of papaya in the prevention and treatment of obesity and associated metabolic disorders.

References

1. Aravind G, Debjit Bhowmik, Duraivel. S, G H. Traditional and Medicinal Uses of Carica papaya. *Journal of Medicinal Plants Studies* 2013; **1**: 07-15
2. Fan JG, Kim SU, Wong VW. New trends on obesity and NAFLD in Asia. *J Hepatol* 2017; **67**: 862-873 [PMID: 28642059 DOI: 10.1016/j.jhep.2017.06.003]
3. Mitra S, De A, Chowdhury A. Epidemiology of non-alcoholic and alcoholic fatty liver diseases. *Transl Gastroenterol Hepatol* 2020; **5**: 16 [PMID: 32258520 DOI: 10.21037/tgh.2019.09.08]
4. O'Hare TJ, Williams DJ. Papaya as a Medicinal Plant. In: Ming R, Moore PH. Genetics and Genomics of Papaya. New York, NY: Springer New York, 2014: 391-407
5. Santana LF, Inada AC, Espirito Santo BLSd, Filiú WFO, Pott A, Alves FM, Guimarães RdCA, Freitas KdC, Hiane PA. Nutraceutical Potential of Carica papaya in Metabolic Syndrome. *Nutrients* 2019; **11**: 1608 [PMID: 31315213 DOI: 10.3390/nu11071608]
6. Tridge C, LTD. Papaya global import and top importing countries Tridge 2020. Available from: <https://www.tridge.com/intelligences/papaya/import>.

7. Younossi ZM. Non-alcoholic fatty liver disease - A global public health perspective. *Journal of Hepatology* 2019; **70**: 531-544 [DOI: 10.1016/j.jhep.2018.10.033]

Comment 2 In the result section "Effects of papaya on lipid accumulation". There are many repeated sentences from the method section. The result is only used to report the result.

Response: We corrected as comments.

Comment 3 Figure 1, row 2. The oil-red O stain is not convincing, even normal liver, the oil-red-O can show few lipid droplets. I would recommend to add one more row of higher power picture to show the red lipid droplets clearly.

Response: Thank you for your comments. We corrected Figure 1, row 2 by choosing the higher resolution to show the lipid droplets clearly and we chose the new figure to represent the normal liver which has clearer image.

Comment 4 Still Figure 1, row 3. The H&E stained histology is not convincing. In this power, I do not see clear fat droplets, even in the HFD group. Please add a row of higher power picture. Also I want to make sure you need to show the ballooning degeneration of hepatocytes and lobular inflammation in a higher power picture. Since the HFD group mean NAS score is 6, that means the liver shows NASH. But the histology picture do not show features of NASH. The authors also mentioned that the fibrosis stage (0-4) was evaluated separately from NAS. How did the authors evaluate it? any special stains used? The authors also said "The experimental groups were blinded and evaluated by the pathologist designated from the Faculty of Medicine, Naresuan University." Who is that pathologist? I do not see any pathologist author from the author list? Who took the pictures for this paper? The authors should be clear that the pathology evaluation is very important and critical for this current study. If there are no pathologic changes among these groups, other experimental data will not be convincing.

Response: We chose the clearer picture that state ballooning and inflammation as suggested as seen in the figure in the revised MS.

Normally, fibrosis can be evaluated using H&E. In this study, we evaluated the NASH from NAS score and found the appearance of hepatic ballooning and inflammation which is a dominant characteristics of hepatic steatosis. No fibrosis was observed at this stage. Others stated that the prolong treatment of HF diet for 16 to 80 weeks can induce liver fibrosis (Kohli, et al., 2010; Velázquez, et al., 2019). For our study, we had a shorter period of 12 weeks.

Anyway, we corrected the content to make it clear as seen in the revised MS. For evaluation, the first author (WD) performed the analysis under supervised from IP (co-author) who has been worked in the histology field for almost 20 years. We also got helpful suggestions and technical advice for NASH evaluation from the pathologist who we acknowledged in the acknowledgement section. From the result figure as shown in the revised MS, it is suggested that there are the pathological changes observed in the HF group when compared to the C group.

References:

1. Kohli R, Kirby M, Xanthakos SA, Softic S, Feldstein AE, Saxena V, Tang PH, Miles L, Miles MV, Balistreri WF, Woods SC, Seeley RJ. High-fructose, medium chain trans fat diet induces liver fibrosis and elevates plasma coenzyme Q9 in a novel murine model of obesity

and nonalcoholic steatohepatitis. *Hepatology* 2010; **52**: 934-944 [DOI: <https://doi.org/10.1002/hep.23797>]

2.Velázquez KT, Enos RT, Bader JE, Sougiannis AT, Carson MS, Chatzistamou I, Carson JA, Nagarkatti PS, Nagarkatti M, Murphy EA. Prolonged high-fat-diet feeding promotes non-alcoholic fatty liver disease and alters gut microbiota in mice. *World journal of hepatology* 2019; **11**: 619-637 [PMID: 31528245 DOI: 10.4254/wjh.v11.i8.619]

Reviewer 2: I think it is a good study showing the beneficial effects of papaya.

Response: Thank you for your support.

2. Editorial Office's comments

Science Editor:

Comment 1 Scientific quality: The manuscript describes a basic study of the papaya improves nonalcoholic fatty liver disease in obese rats by attenuating oxidative stress, inflammation and lipogenic gene expression. The topic is within the scope of the WJG. However, the scientific quality doesn't meet the requirement of WJG. (1) Classification: Grade D and Grade D;

Response: -

Comment 2 Summary of the Peer-Review Report: The authors tried evaluate the anti-NAFLD effect of papaya in high fat diet induced obesity in rats, and concluded that papaya can suppress the lipogenic pathway, improve the balance of antioxidant status and lower systemic inflammation. The study is well designed. It is a good study showing the beneficial effects of papaya. However, the authors need to review the literature and discuss any health benefits in general population and individuals with obese and other health issues. Some repeated sentences from the method section should be deleted and revised. The questions raised by the reviewer should be answered

Response: We corrected the contents as suggested by the reviewers.

Comment 3 Format: There are 2 tables and 5 figures. A total of 34 references are cited, including 3 references published in the last 3 years. There are no self-citations. 2 Language evaluation: Classification: Grade A and Grade B. 3 Academic norms and rules: The authors provided the Biostatistics Review Certificate, the Institutional Animal Care and Use Committee Approval Form or Document, and The ARRIVE Guidelines. The authors should provide the signed Conflict-of-Interest Disclosure Form and Copyright License Agreement. No academic misconduct was found in the Crosscheck detection and Bing search.

Response: We attached the signed conflict of interest form and the copy right license agreement for the revised MS.

Comment 4. Supplementary comments: This is an unsolicited manuscript. The study was supported by The National Research Council of Thailand, and The Thailand Research Fund. The topic has not previously been published in the WJG.

Response: -

Comment 5 Issues raised:

5.1 The authors did not provide the approved grant application form(s). Please upload the approved grant application form(s) or funding agency copy of any approval document(s);

Response: We prepare to upload the approved grant documents.

5.2 The authors did not provide original pictures. Please provide the original figure documents. Please prepare and arrange the figures using PowerPoint to ensure that all graphs or arrows or text portions can be reprocessed by the editor; and

Response: We prepared the figures using PowerPoint as recommended.

5.3 The “Article Highlights” section is missing. Please add the “Article Highlights” section at the end of the main text.

Response: We added the article highlights at the end of the main MS.

Comment 6. Re-Review: Required.

Response: We submitted the revised MS for re-review.

Comment 7 Recommendation: Suggest to transferring to World Journal of Hepatology.

Response: Thanks for the opportunity to transfer our MS to the World Journal of Hepatology.

2. Editorial Office Director: I have checked the comments written by the science editor.

Response: -

3. Company Editor-in-Chief: I recommend the manuscript to be published in the World Journal of Hepatology

Response: Thank you for the consideration to publish our MS to the World Journal of Hepatology

Round 2

Thank you for the comment. We replaced the histological picture with the same magnification as others as suggested. The correction was seen in Fig1, row 3 for the HFD group.