

Tuesday, February 11, 2014



Dears Editors

Title: The beneficial effect of butyrate, Lactobacillus casei and L-Carnitine combination in preference to each in experimental colitis

Name of Journal: *World Journal of Gastroenterology*

ESPS Manuscript Number: 7078

Invitation ID: 00040588

Thank you and the respected reviewers for very positive and helpful comments. We have considered all points very carefully and did our real best and revised the paper very thoroughly. We have kept the revised in track change mode to make you know the extent of our revision and caution. In the meantime, we have described below whether we have dealt with each comment or recommendation of reviewers.

Editorial

- 1- Format has been updated to be in full adherence to WJG.
- 2- Revision has been made according to the suggestions of the reviewer
- 3- Thorough English language editing is made either by Corresponding author who an English Language educated academic and a native from UK whom her name has been acknowledged.

Reviewer 1

(1) This work is both very interesting and timely research. The implications for IBD management are fascinating, particularly taking into account the potential for butyrate, with its possible antineoplastic action, as a treatment for condition with an increased cancer risk. The introduction reads well but the first sentence should be reviewed as this does not appear to be a correct description of IBD.

Done as recommended; The first sentence changed to: Inflammatory bowel disease (IBD), representing as ulcerative colitis (UC) or Crohn's disease (CD), with an increasing incidence, can be debilitating in affected patients. In this part we didn't want to go to details as IBD is a well-known disease because for readers of a GI specific journal like WJG. Therefore, we tried to focus on conventional therapies and the patients' incomppliance to justify the need for introducing new effective therapies with lower toxicities.

(2) The method is very thorough and well described. It may helpful to state exactly how long after the TNBS that treatment was given.

Corrected and added to the methods.

(3) At times throughout the manuscript you vary between UC and TNBS induced colitis, the second description seems more accurate.

We agree with the comment and thus named it "colitis" instead of UC.

(4) Also in the list of equipment, company names are given for some but not others, if possible this should be more consistent.

Now they are consistent.

(5) The results are quite difficult to read but the figures are very helpful and clear. In the discussion, the mices' stool consistency is mentioned which does not appear to have be measured and is not described elsewhere.

We have done a huge edit on Results section to mention all changes in comparison with Sham, Control, Positive control, Combination, and Single therapies with P values. Now everybody by reading the Results can understand what are the changes.

(6) Overall, whilst this very interesting and complete work, this paper would benefit from a thorough review of the language used as some sentences do not seem to make sense and it appears that the wrong word has been used in some instances.

Linguistic edit has been done twice by an English-language educated academic and a native academic. Now it is okay, we guarantee.

Reviewer 2

(1) Abstract: Please improve the language grammatically. Conclusion needs to be rewritten.
Done thoroughly as mentioned above too. Conclusion has been added.

(2) Introduction: Needs to be improved
Now improved as described above for the reviewer 1.

(3) Materials and methods: Each and every sentence needs to be reworded.
Corrected. We have put “and” instead of “comma”.

(4) Probiotics is a wide term and can be used in general context not in research paper. Since, heath benefits exhibited by probiotics are strain specific, the name of the strain used should be given along with source and any particular scientific information available should be mentioned in the paper. The authors have only given under source that it was procured as a powder of *Lactobacillus casei* from Zist-Takhmir Co (Tehran, Iran).

Corrected; We replaced the probiotic with “*L. casei*” as suggested in whole article. The name of strain used in our lab (*L. casei* DN: 114001; lab no: 9802) was added.

Did they authenticate the species themselves in their laboratory?
Yes, the seller Zist-Takhmir company produces several strains of bacteria including *L. casei*.

Is this strain commercially available and any strain number assigned to this particular *L. casei*?
Yes, the producer company provides such strains of *L. casei*. Biochemical and molecular tests are used to identify our desired type of *L. casei*.

Probiotic word should be replaced with *L. casei* (also indicate strain) throughout the manuscript along with Title.
Corrected everywhere.

What is the total viable cell count in ¼ teaspoon of *Lactobacillus casei*? Replace the sentence indicating cell number. How the cell number of 1×10^8 was adjusted each time?
The sentence was reworded for clarification; We utilized count test before any experiment to make sure the cell number is 1×10^8

(5) Results: Very poorly described. Data should always be presented by giving values and percent reduction which will be more clear in comparison to writing significant increase or decrease in activity. Comparative evaluation of different agents can only made by quantitative description of data.

As we explained in respond to the Reviewer 1, we have totally edited the Results part. Within the text, percentage changes and the p values were added. All changes in comparison with Sham, Control, Positive control, Combination, and Single therapies with P values have been noted. Now everybody by reading the Results can understand what are the changes.

Macroscopic and microscopic evaluation of histological impairment

Table 1 and 2 indicating scoring of colonic damage should be taken into supplementary information.

Thanks for this comment; We in fact removed the tables 1 and 2, and thus described the scoring system as text in the methods.

Table 3: Please make the following corrections.

Complete the word “Dexa” Replace probiotics with “*L. casei*” and correct L. carnitin in both Table 1 and Fig.1

All done.

Results to be described in quantitative terms giving values from Tables and Figures not just writing increased or decreased activity

Done but that was just for reducing the paper length. We cannot bring all details as we address tables and figure for that purpose. But this comment of review is met now.

MPO activity

Give the values for MPO activity and replace significant reduction also with quantitative data.

Done.

TNF- α level and IL-1 β level, Anti-oxidant power as FRAP and Oxidative-stress as TBARS

Please rewrite again giving values.

Done

Discussion

Discussion also needs to be rewritten giving proper explanations of the results obtained. The authors have given general statements which are well known and have not discussed why the combination therapy is better than the effect induced by individual agents. Some of the parameters are better in case of individual agents as compared to combination. What is the explanation for those?

Overall results are in favor of combination especially in the critical markers of oxidative stress such as TBARS, FRAP, and MPO. We have revised discussion to meet this comment and to be clear in mentioning the statistical differences and why combination group can be considered almost better than single therapies. Anyway we have reworded this part to meet the reviewer point.

The authors have not explained the proposed mechanism by the combination treatment as shown in Figure 7 anywhere in the paper. This hypothesis has been published in their previous paper. The authors should have validated the proposed mechanism in their paper which would have enhanced the quality of their paper. Nowhere in the paper, reference to Fig. 7 has been made, hence should be deleted by giving reference of previous paper.

More description was added addressing the reference. In fact, our idea to make this combination effective in practice has been already published in Journal of Medical Hypotheses and Ideas. With citing that paper and the Figure 7 adapted from that OA source, we have explained the mechanisms by which each component of the mixture plays role in ameliorating the colitis.

Conclusion: Needs to be rewritten

Done

3 References and typesetting were corrected

Thank you again.