

Date: April 4, 2017

Dear Professor Ma,

We would like to thank you for giving us the opportunity to revise our manuscript entitled “Dietary and metabolomic *determinants of relapse in ulcerative colitis patients: a pilot prospective cohort study*” for possible publication in World Journal of Gastroenterology. Please find our point-by-point reply to all comments from the reviewers in the second page of this letter. All relevant changes have been highlighted in yellow color in the revised manuscript.

Please kindly notice that this manuscript was previously submitted as an invited manuscript (Author ID: 03257654, manuscript NO. 32615). However, the manuscript got rejected and we decided to resubmit the manuscript after revising it based on the comments from the reviewers. We sincerely hope that these changes have addressed the comments to your satisfaction. We also hope that you can waive the publication fee for this manuscript and still consider it as an “invited manuscript”. Thank you very much for your consideration.

Yours sincerely,

Ammar Hassanzadeh Keshteli, MD

On behalf of all co-authors

Answer to the comments:

- **Reviewer's code: 00068574**

Comment: Minor point Page 8, line 34. Please add "In" at the beginning of the sentence: "Another recent study"

Answer: "In" has been added to the revised manuscript.

- **Reviewer's code: 00049433**

Comment: Intro: please add an aim/hypothesis to the end of the introduction

Answer: The last paragraph of the introduction section has been edited according to your comment.

Comment: Is a Mayo <3 enough of an inclusion criteria?

Answer: We think this is an enough inclusion criteria for "clinical remission". Please see reference #17.

Comment: Is two weeks off steroids enough to stay that the patient is truly in clinical remission?

Answer: We have used a combination of several factors to make sure our patients were in "clinical remission" at the time of enrollment. If our aim was to include patients in "endoscopic remission" or with mucosal healing for sure this inclusion criteria (partial Mayo score less than 3 or criteria regarding medication) was not enough.

Comment: Were eligible patients recruited consecutively - if not then significant selection bias may have occurred

Answer: Thanks for your comment. Yes, they were recruited consecutively. This has been added to the methods section of the revised manuscript.

Comment: Although the FFQ is validated in cancer patients, has it been validated in IBD? And is there not likely to be recall issues when asking people about their diet over a 12 month period?

Answer: The NCI FFQ we used in this study has not been designed for using only in Cancer patients and so far it has been used in several settings. It is a widely used questionnaire and has been shown to be a valid tool for assessment of dietary intake among "adult" population. However, we agree with you that FFQ is subject to recall bias. We have added this to the limitation paragraph of the discussion section.

Comment: Although this is clearly a pilot project I have some concerns about the conclusions drawn on these numbers. Given the number of hypotheses this study was testing, were all p values corrected for multiple testing (I don't believe so reading the manuscript). I suspect if not,

then many of these "positive" results may not be truly positive. For example testing FCP, weight, medications, metabolites etc on only 20 patients is very likely to produce false positive results. I think a power calculation for this study is required, if only to show the reader that this pilot is underpowered.

Answer: Thanks for your thoughtful comment. We agree with you that the multiple testing might have been a source of false positive findings in this study. We have tried to highlight this in the limitation paragraph of the discussion section. Please also note that the statistical tests we used for the metabolomic data are robust techniques and use a multivariate type of statistical analysis to reduce chance of false positive findings. As you know "power is the probability of rejecting a null hypothesis when it's false". Therefore, power of a study does not deal with "false positive" findings and it is usually calculated when you report some non-significant P-values (false negative findings).

Comment: Discussion: this seems very long and needs significantly shortened

Answer: However, since we investigated a lot of potential determinants of relapse in this study, we had to have at least one paragraph for each of these factors which have resulted in a relatively long discussion section. However, in the revised manuscript we have shortened some paragraphs as you suggested.

- **Reviewer's code: 03262333**

Comment: As this is a pilot study the sample size and the recruitment strategy are not well described It is not clear from how many patients this group was selected It is interesting no one in this group did not use biologic treatment

Answer: We did not perform a sample size calculation for this pilot study as the aim was to check the feasibility of the study in our center and identification of potential contributors of clinical relapse. No patient in this study was on biologic agents as this was an exclusion criteria for this study.

Comment: The baseline data on clinical and endoscopic severity index were not provided

Answer: In Table 1, we have provided several clinical data at baseline and as described in the methods section of the manuscript our focus was on "clinical remission" which is based on partial Mayo scoring and does not require endoscopic evaluation.

Comment: The multi regression analysis is not provided. For instance it seems older, overweight were protected from relapse. Which one was the major factor?

Answer: As clearly indicated in Table 1, there was no relationship between age and UC clinical relapse. However, as you suggested, in the revised manuscript we have presented P-value for the relationship between overweight/obesity after adjusting for age and gender and added relevant information to the methods and results section of the manuscript.