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Dear editor,

Thank you very much for your attention and the reviewers' evaluation and comments on our paper. Your comments and those of the reviewers were highly insightful and enabled us to greatly improve the quality of our manuscript. We have revised the manuscript according to your kind advices and reviewers' detailed suggestions. The revised parts are highlighted in the manuscript. Enclosed please find the responses to the referees. We sincerely hope this manuscript will be finally acceptable to be published on World Journal of Gastroenterology.

We shall look forward to hearing from you at your earliest convenience.

Yours sincerely,

Jia-dong Zheng, Harbin Medical University, 157 Baojian Rd, Harbin,China.

Tel:+8618343228229

E-mail: zjiadong@yeah.net

Responses to the comments of Reviewer 1

Reviewer's code: 04025483

1. Please clearly indicate the number mice used for every experiment and add this information to the respective figures.

   Response: Thanks for the referee's suggestion. We have revised the details in the MATERIALS AND METHODS of the manuscript in Page8, line 204-206. And we have revised the figure legend of every figure.

2. In addition to measuring serum D-lactate, did the authors also check for DAO? In addition to these indirect evidence, the authors should also make attempts to directly visualize restoration of the intestinal barrier e.g. via the use of fluorescently labeled dextran.

   Response: Thanks for the referee's suggestion. Just like what the referee said the use of fluorescently is more directly visualization. However, due to the shortage of experimental samples we test serum D-lactate and occluding level of colon to indicate the change of intestinal barrier. So we didn't test DAO. If this is necessary, we will try our best to remedy the shortage of this experiment. And in subsequent research on UCB or IBD, we will add fluorescently to test the intestinal barrier function.

3. Why was UCB administered via Gavage and not by enema? Also, why was this dose chosen? Did the authors perform dosing curves prior to the experiments that are actually shown?

   Response: Thanks for the referee's question. We have also thought about this question. The research purpose of our team is to explore novel drugs for IBD. Oral administration is more convenient for people. And in previous studies on IBD-CD, our team has been using intragastric administration to simulate oral administration in humans. About the dose of UCB, we performed UCB at concentrations of 100,200,400,600 uM, respectively. The preliminary experiments found 400 μM UCB could significantly improve UC mice as shown in Figure 1.

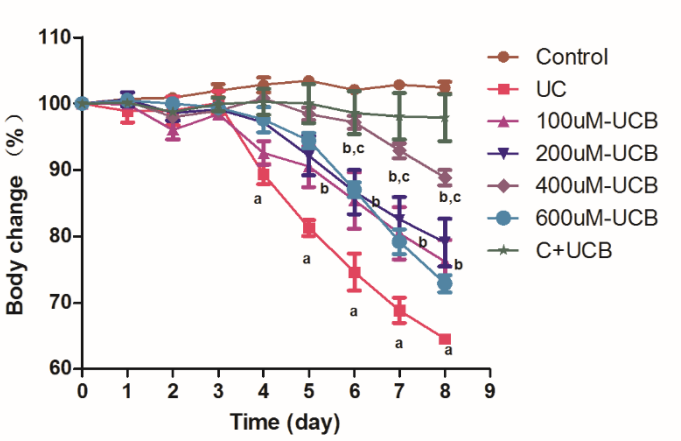

Figure 1. Body change. ^{a}P: < 0.001 vs control, ^{b}P: < 0.001 vs UC, ^{c}P: < 0.001 vs 100Mm UCB,200μM UCB and 600Mm UCB.

4. The authors should also analyze the role of UCB in chronic DSS colitis. If they could show that UCB can ameliorate also chronic DSS and eventually carcinogenesis (AOM-DSS model), this would tremendously increase the significance of their findings.

   Response: Thanks for referee's kind advice. Our team is also establishing the chronic DSS colitis and inflammatory colon cancer models. And the next research will be the role of UCB in chronic DSS colitis and colon cancer.

5. The authors state that they have shown in prior studies that UCB can ameliorate the inflammation in trinitrobenzenesulfonic acid (TNBS)-induced colitis. However, the reference provided here is not correct. Please check and provide the correct reference.

   Response: Thank you very much for referee's comment. We have revised it in Page 13, Line 362 of the manuscript. And the prior study article is **Zhou JA**, Jiang M, Yang X, Liu Y, Guo J, Zheng J, Qu Y, Song Y, Li R, Qin X, Wang X. Unconjugated bilirubin ameliorates the inflammation and digestive protease increase in TNBS-induced colitis. *Mol Med Rep* 2017; **16**(2): 1779-1784.

6. Minor concerns: 1) Figure 1 C: please add labeling to indicate which colon belongs to which treatment group. 2) Language polishing both in style and

grammar is necessary prior to publication

Response: Thanks for referee's comment. The revised detail can be found in Page 21 Figure 1C. The manuscript has been edited for English language

by a native English speaking medical editor at MedE Medical Editing Group.

Responses to the comments of Reviewer 2

Reviewer's code: 02821831

1. The authors must add in introduction the involvement of Pro Inflammtory cytokine and Nitric oxide in inflammatory procces in IBD (Rafa etal, 2013, Soufli et al,2016) The eventual modulation of inflammatory pathway (TL4/NFKB) by probiotic agents needs some attention by the authors(Toumi et al, 2014).

   Response: Thanks for referee's kind advice. We added this point into our revised manuscript and the details can be found in Page 6, Line 147-151 and Line 153-157.

2. The clinical relevance of the study must be added in section Discussion.

   Response: Thanks for referee's kind advice. We added this point into our revised manuscript and the details can be found in LPage14-15, Line 407-414.

Responses to the comments of Reviewer 3

Reviewer's code: 03479673

1. How many mice were in each group?

   Response: Thanks for referee's question. Formal Laboratory, 5 mice were in each group.

2. How did you randomize these mice to particular group? And at what timeline of study randomization was done?

   Response: Thanks for referee's advice. We purchased SPF C57BL/6 male mice from the animal experimental center, they were fed in a standard condition for one week to adapt the environment, and then they were weighed and randomly divided into four experimental group as Control, UC，UCB+UC and UCB with 5 mice every group on the Day0. UC and UC+UCB group drunk 3% Dextran Sulfate Sodium Salt (DSS) in water for 6 days, and then filtered water for 2 days. These were 8 days. UCB was administered via gavage at 400 μM, 0.2 mL from Day 1 to Day 7. UC+UCB and UCB group mice had intragastric administration for 7 times. Finally, all mice were sacrificed on the Day8 after weighing.

3. What was the calculated sample size you derived from the statistical calculation? And how did you calculate the same?

   Response: Thanks for referee's advice. Due to the the ethical protection of animals, the number of animals is limited. The calculated sample size derived from the statistical calculation of histopathological staining and western blot was 3 from different mice. The calculated sample size of other experiments was 5 from different mice. All experiments were repeated three times. Results were expressed as mean ± SEM. Differences between groups were determined using one-way ANOVA followed by Tamhane multiple comparisons post-hoc tests using SPSS version 19.0. Statistical significance was denoted with P values < 0.05.

4. Blinding of the analyst was done or not?

   Response: Thanks for referee's advice. Every author of the

manuscript has his or her own contribution. Jia-dong Zheng is responsible for analysis and interpretation of data, and writing the manuscript as the first author of the manuscript, and Yan He, Heng-yuan Yu, Yi-xuan Ge, and other colleague.

5. Were the mice in all groups matching in their characteristics before starting the study intervention?

   Response: Thanks for referee's question. The mice in all groups were matching in their characteristics before starting the study intervention. The mice were purchased at the laboratory animal center a week before the experiment started. All mice were 8-12 week male C57BL/6 mice (weight ~25g).