Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 7926R1.docx).

Title: Mast cell deficiency exacerbates inflammatory bowel symptoms in IL-10-deficient mice

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Name of Journal: *World Journal of Gastroenterology*

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The manuscript has been improved according to the suggestions of reviewers:

Reviewer#1:

In this study, the authors cross-bred mast cell-deficient mice with IL-10-deficient mice to investigate the role of mast cells in gut inflammation and the onset of colitis. Data show that mast cells have protective roles in the development of colitis by suppressing Th1 type immune response and inflammation, altering gut microbiota composition, improving gut epithelial barrier function, and reducing epithelial damage. REMARKS. I suggest to show the histological pictures present in Figure 1 at higher magnification.

Response: Thank you. We prefer to show the whole gut cross-section structure because the pathological score was given based on the whole structure. Due to the heterogeneity of inflammation, arbitrary selection of a small section of the gut with/without inflammation might introduce bias to the results.

Reviewer#2

The authors are describing very interesting results about the effects of mast cell deficiency on colitis in a double knockout mouse model obtained by cross-breeding mast cell-deficient mice with IL-10-deficient mice. The paper is well-written, the methods used are sound, the results are novel and are a basis for future work in this research area.

I recommend to accept this paper for publication in the World Journal of Gastroenterology as it stands.

Response: Thank you.

Reviewer#3

This manuscript examines colitis development in IL-10 deficient mice when crossed with mice carrying the c-kit W "sash" mutation. This is thus similar to a study published in 2010 by different authors showing that these double mutants develop enhanced colitis and intestinal permeability. This manuscript is thus confirmatory but does provide some new data.

1. There are some new data presented, but they add little to our understanding of how mast cells normally reduce severity of IBD in IL-10 deficient mice.
   Response: Based on our observation that systemic inflammation was detected, we proposed that systemic inflammation due to the double deficiency in IL-10 and mast cells might be the cause for the enhanced IBD severity, which is novel.

2. Changes in claudins 2 and 3 re reported. Claudin2 mRNA is unchanged but protein is increased. Claudin 3 mRNA is decreased but protein is not. The authors must address the discrepancies between their mRNA and protein data.
   Response: The discrepancies should be due to translational regulation. The enhanced inflammation in DKO mice likely generates translational regulation. There are a number of studies show that inflammation is able to elicit changes in translational controls (Mazumder et al., 2010; Novotny et al., 2012).

3. A change in Rhamnococcus abundance is reported, but it is unclear how this relates to the development of colitis in this model.
   Response: Our observation that Ruminococcus was increased is consistent with previous reports showing that it is Inkreise in patients with inflammatory bowel diseases (Willing et al., 2010)Kang et al., 2010). The reason for the increase of this particular bacterial species is unclear, but could be related to the overall changes in gut microbiota due to IBD. Very recent studies emphasize the importance of overall changes in microbial composition, or microbiota and gut metabolomics to gut health.

4. How does the high energy diet in the mothers effect these results? It is odd that control diets did not produce successful breeding. The data on glucose metabolism and body fat are not well connected to a role for mast cells in the context of IBD and may be a completely separate issue.

Response: Yes, we agree with the reviewer. It was unexpected but the case. High fat diet might rescue the deficiency in fertility due to mast cell deficiency, but the exact reason is unclear, which is beyond the scope of current study.

Minor comments.

1. The authors should not refer to double knockouts. Sash is a hypomorphic allele of Kit, not a knockout.
   Response: Thanks for your comments. We understand that it is due to a mutation not KO. However, these mice are deficient in mast cells. For simplicity, we call it mast cell KO. We have provided additional explanatioin.

2. Please be consistent in use of FFA (not FAA). Why do methods refer to fetal plasma fatty acids??
   Response: This has been corrected.

3. GAPDH is misspelled in table 2
4. **Response:** This has been corrected. Thanks!

References:

Kang, S. et al. 2010. Dysbiosis of fecal microbiota in crohn's disease patients as revealed by a custom phylogenetic microarray. Inflamm Bowel Dis 16: 2034-2042.

Mazumder, B., X. Li, and S. Barik. 2010. Translation control: A multifaceted regulator of inflammatory response. Journal of immunology 184: 3311-3319.

Novotny, G. W. et al. 2012. Transcriptional and translational regulation of cytokine signaling in inflammatory beta-cell dysfunction and apoptosis. Archives of biochemistry and biophysics 528: 171-184.

Willing, B. P. et al. 2010. A pyrosequencing study in twins shows that gastrointestinal microbial profiles vary with inflammatory bowel disease phenotypes. Gastroenterology 139: 1844-1854 e1841.