Dear Editor and Reviewer:

We would like to thank you and your reviewers for taking the time to consider our manuscript (ESPS Manuscript NO: 32393) and for providing constructive criticism. Here we submit our revised manuscript, with tracked changes for ease of comparison. In addition, please find below our responses to the individual comments made by the reviewers.

We believe that the peer review process has assisted us in producing a significantly improved manuscript, and we look forward to your assessment of our re-submission to WJG.

Reviewer comments:

Comment 1： *Figure 3 shows that BMMSCs transduces with the adenovirus CXCR3/HO-1 construct express both CXCR3 mRNA and protein. However, the levels of HO-1 mRNA and protein do not appear to have been evaluated. Concurrent evaluation of both HO-1 and CXCR3 mRNA and protein levels at this step would be helpful.*

Response: Thank you very much for your valuable suggestion. We have made appropriate amendments in the revised manuscript.

Comment 2： *Figure 4 shows histologic evaluation of small bowel after transplant for each of the experimental groups. The quality of the figure is too poor to properly evaluate the author's conclusions. Further, the materials and methods describe using a histologic method from 1970 to describe "pathologic rejection." If the authors want to make a claim regarding the effect of the modified BMMCSs on acute rejection, a more modern grading scheme should be used such as "Transplantation. 75(8):1241-1248, April 27, 2003." While I cannot evaluate the histology due to image quality, the description of the pathologic changes does not mention any vascular findings such as arteritis. Nor does the description mention any cytologic changes such as apoptosis (later evaluated by fluorescence, but traditional grading of rejection*

relies on H&E) or nuclear enlargement and/or hyperchromasia in the epithelium. Are the histologic changes observed due to ischemic issues in the BM group? How patchy or confluent were the described lesions? What type of inflammatory infiltrate is increased (acute or mononuclear cell?)?

Response: Thank you very much for your valuable suggestion. The classification of pathological rejection in this paper has been made revised in the second iteration of the manuscript according to your suggestion [1]. Pathological changes within the vascular system, crypt epithelial cells and lymphocyte infiltration have all been described in the revised manuscript. Histological changes in the NS group were caused by ischemia-reperfusion injury. However, the histological changes in the BM group were mild because BMMSCs can repair them. We have changed the description of the experimental group in response to the reviewer comment. The BM group, HB group, and HCB group were compared with the NS group, respectively. We found that infiltration of inflammatory cells was primarily due to mononuclear cells.

References:

1 **Wu T**, Abu-Elmagd K, Bond G, Nalesnik MA, Randhawa P, Demetris AJ. A schema for histologic grading of small intestine allograft acute rejection. *Transplantation* 2003; **75**(8): 1241-1248.

Comment 3： Figure 6 B shows the same chi-squared and P value for every comparison. Can this be correct?

Response: Yes, it is correct. We have confirmed that the chi-square and P values for each group are the same.

Comment 4： *Figure 7 is not clear as to the source of cells or tissue analyzed. Did the authors remove a piece of intestine from the rats on days 1, 3, 7, 10 and 14 and analyze the protein expression on that tissue? If so, the figure caption should reflect that.*

Response: Thank you very much for your suggestion. The protein shown in Figure 7 is derived from the small intestine tissue from the rats on days 1, 3, 7, 10 and 14. We have made appropriate amendments in the revised figures.

Comment 5: *Does manipulating the transplanted bowel tissue so invasively have any potential to bias the results? Why is this a viable strategy rather than sacrificing rats at a certain time point and comparing the same expression patterns then?*

Response: Thank you for this valuable suggestion. The results are not significantly affected by bias because we performed small bowel transplantation on the recipient rats using the same experimenter. Furthermore, we used a unified arrangement for the control of abiotic factors e.g. experiments were conducted in the same room temperature and air environment. Moreover, the weight of rats, the length of the transplanted small intestine, the time of operation, the cold ischemia time of the transplanted small intestine and other parameters were all the same. In addition, the control group (NS group) was set up at the same time as the experimental group, therefore we were able to remove the confounding effects of surgery itself.

Our experiment aimed to sacrifice the recipient rats simultaneously, which was consistent with your point of view, but we divided the study into six time points for experimental comparisons. Our study was based on the chemotaxis of CXCR3 to explore whether CXCR3-modified BMMSCs could reach the site of inflammatory injury faster and as a more enriched population of BMMSCs. Our study sacrificed the recipient rats at the same time point and we observed the expression of the recipient rats at the same time point. The rejection response caused by the inflammatory response increases gradually during the process, with rejected CXCR3 ligand also increasing [2-4]. Therefore, we chose to sacrifice rats at multiple time points to describe the results in further detail. We do not believe that selection of a point in time can reflect this. We have made appropriate amendments in the discussion section

of the revised manuscript.

References:

2 **Shino MY**, Weigt SS, Li N, Palchevskiy V, Derhovanessian A, Saggar R, Sayah DM, Gregson AL, Fishbein MC, Ardehali A, Ross DJ, Lynch JR, Elashoff RM, Belperio JA. CXCR3 ligands are associated with the continuum of diffuse alveolar damage to chronic lung allograft dysfunction. *Am J Respir Crit Care Med* 2013; **188**(9): 1117-1125.

3 **Hricik DE**, Nickerson P, Formica RN, Poggio ED, Rush D, Newell KA, Goebel J, Gibson IW, Fairchild RL, Riggs M, Spain K, Ikle D, Bridges ND, Heeger PS. Multicenter validation of urinary CXCL9 as a risk-stratifying biomarker for kidney transplant injury. *Am J Transplant* 2013; **13**(10): 2634-2644.

4 **Pranzatelli MR**, Tate ED, McGee NR, Travelstead AL, Verhulst SJ, Ransohoff RM. Expression of CXCR3 and its ligands CXCL9, -10 and -11 in paediatric opsoclonus-myoclonus syndrome. *Clin Exp Immunol* 2013; **172**(3): 427-436.

Comment 6：*Figure 9 shows expression of CXCR3 and HO-1 protein in the small bowel. What cells are likely being stained? Are these actually BMMCSs?*

Response: Thank you very much for your comment. Chemokine receptor CXCR3 is expressed in inflammatory cells such as vascular endothelial cells, activated lymphocytes, macrophages and dendritic cells *in vivo*. However, it is not expressed in quiescent T lymphocytes, B lymphocytes and granulocytes [5, 6]. HO-1 is mainly distributed in the tissues and organs of blood cell-rich organs, such as the spleen, liver and bone marrow; HO-1 expression is very low in other tissues [7]. Thus, CXCR3 protein and HO-1 protein co-expression in the same cell is very unlikely. Given that there are two proteins in CXCR3/HO-1/BMMSCs, when these cells reach the damaged site, double protein expression will occur at the same time. This results in a dual fluorescence phenomenon, with fluorescing cells being BMMCSs.

References:

5 **Garcia-Lopez MA**, Sanchez-Madrid F, Rodriguez-Frade JM, Mellado M, Acevedo A, Garcia MI, Albar JP, Martinez C, Marazuela M. CXCR3 chemokine receptor distribution in normal and inflamed tissues: expression on activated lymphocytes, endothelial cells, and dendritic cells. *Lab Invest* 2001; **81**(3): 409-418.

6 **Huang H**, Xu X, Yao C, Cai M, Qian Y, Wang X, Shi B. Serum levels of CXCR3 ligands predict T cell-mediated acute rejection after kidney transplantation. *Mol Med Rep* 2014; **9**(1): 45-50.

7 **Marcantoni E**, Di Francesco L, Dovizio M, Bruno A, Patrignani P. Novel insights into the vasoprotective role of heme oxygenase-1. *Int J Hypertens* 2012; **2012**: 127910.

With best wishes

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