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Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 10346-Review)

Title: Alleviated mucosal and neuronal damage in a rat model of Crohn's disease

Authors: Petra Talapka, Lajos István Nagy, Alexandra Pál, Marietta Zita Poles, Anikó Berkó, Mária Bagyánszki, László Géza Puskás, Éva Fekete, Nikolett Bódi

Name of Journal: *World Journal of Gastroenterology*

ESPS Manuscript NO: 10346

Answer to Reviewer 1 (in order of paragraphs)

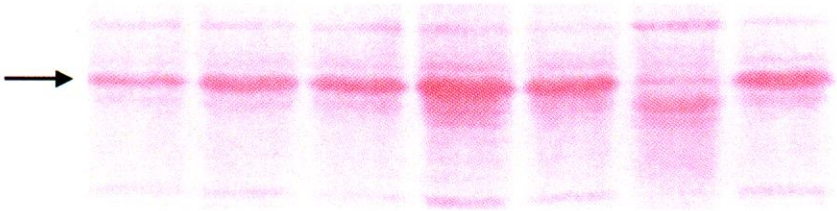
1/ Thank you for reviewing our manuscript and for your suggestions. Considering your remarks the *Introduction* part has been shortened substantially in the revised version of the manuscript.

2/ Considering your comments on *Figure 4*, relative heme oxygenase-1 (HO-1) mRNA expression level was considered one in each tissue samples derived from the control rats, and HO-1 gene expression was compared to the controls in each TNBS-treated experimental group. Therefore, HO-1 mRNA level in controls is presented as means without SD. When the amount of HO-1 coding mRNA in samples derived from the TNBS-treated groups was higher than one, HO-1 gene expression was regarded as upregulated and when lower than one was regarded as downregulated. In the fourth paragraph of the Discussion part we give some explanation concerning the decrease in HO-1 mRNA levels in proximal and distal colonic segments.

3/ To increase reliability of the experimental data, not only Hypoxanthine guanine phosphoribosyltransferase (HPRT), but also Glyceraldehyde-3-phosphate dehydrogenase (GAPDH; NBCI RefSeq Database entry: NM_017008.4; forward: 5' tgggaagctggtcatcaac 3' and reverse 5' gcatcaccccatttgatgtt 3') were always used through qRT-PCR experiments as housekeeping genes to confirm HO-1 mRNA expression. Since HPRT threshold cycle (Ct) values were more stable, it has been chosen as an internal control to demonstrate differences in relative HO-1 mRNA expression level in samples derived from the control and the TNBS-treated experimental groups *(Figure 4)*.

4/ We naturally agree that the up-regulated expression of HO-1 mRNA which we demonstrated here with real-time polymerase chain reaction does not obviously means an elevated HO-1 protein expression. Considering your advice, therefore we performed western blotting analysis to investigate the changes of the HO-1 protein level (see in *Materials and Methods, Results* and also *Discussion* section of the revised version of the manuscript). We added a new figure with representative gel image prepared from tissue homogenates of the colon of control and TNBS-treated rats (*Figure 5*). Since the molecular weight of beta-actins (approximately 42 kDa) is very close to the molecular weight of HO-1 (32 kDa), equal protein loading was determined by staining the blot with 0.1% Ponceau red in 5% acetic acid (Horváth et al., 2007). Representative gels are shown below, where arrow indicates the loaded proteins. These gels are not shown in the revised version of the manuscript.

Determination of the equal protein loading by Ponceau red staining



We hope that you will accept the revised version of our manuscript and consider it suitable for publication in *World Journal of Gastroenterology*. The changes in the resubmitted manuscript are highlighted by using coloured text.

Answer to Reviewer 2

Thank you for reviewing our manuscript and for your comments and suggestions. Considering your comments the Introduction part has been shortened substantially. The final paragraph of the Introduction section in the revised version of the manuscript includes only the aims of the present study, as you suggested. The Discussion part has been extended with comments about the potential limitations of the model. Considering your advice, we included in the legends to figures the number of animals for each experimental group. We hope that you will accept the revised version of our manuscript and consider it suitable for publication in *World Journal of Gastroenterology*. The changes in the resubmitted manuscript are highlighted by using coloured text.

Sincerely yours,

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