

ANSWERING REVIEWERS

April 17, 2013

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 2188-revised.doc).



Title: The role of *Salmonella enterica* exposure in Chilean Crohn's disease patients.

Author: Manuel Alvarez-Lobos, Daniela P Pizarro, Christian E Palavecino, Abner Espinoza, Valentina P Sebastian, Juan C Alvarado, Patricio Ibañez, Carlos Quintana, Orlando Díaz, Alexis M Kalergis, Susan M Bueno.

Name of Journal: *World Journal of Gastroenterology*
ESPS Manuscript NO: 2188

We appreciate your valuable comments and we are sending a revised manuscript.

Answering to Reviewers' comments:

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

- 1) Format has been updated
- 2) Revision has been made according to the suggestions of the reviewers

(1) Peer-reviewer # 1

This manuscript aimed to study the association between exposure to *Salmonella enteric* and Crohn's disease. In addition, they also analyzed the involvement of cigarette smoking. They showed no association was found in *Salmonella* status and CD, but positive association in smoking and CD. However, the authors should perform following studies in order to address these issues and increase the impact of this manuscript. 1. This paper does not have a novelty because other reports have already similar conclusions. I think it may be valuable if the Chile people have any specific features on IBD or other related diseases. If not, they should analyze more cases to firm the results.

Response : Given the relevant comment made by the reviewer, we emphasize that the value of this study is: 1) to help clarify the real role of *Salmonella* in CD in this controversial subject (page 14, lines 18 to 23), and 2) to confirm previous results in a new and well-defined population (Chilean patients) (page 12, lines 2 to 6 and page 14, lines 18 to 19) . Studies of this disease in our country are very limited so we can not say if our population has some special feature. There are no previous studies on this topic in our country or in South America.

- Regarding the test for *Salmonella* IgG, their sensitivity and specificity should be described.

Response: As requested by the reviewer, we have cited in this new version of the manuscript a study where the sensitivity and specificity of the ELISA assay used in our work was previously evaluated (Mette et al, Clinical and Vaccine Immunology, Vol. 14, No. 6, p. 741-747, 2007). We have modified the manuscript to include this reference (Page 7, lines 20 to 21).

- They showed positive relationship between SE IgG and smoking, this should be analyzed more in detail. There are numerous numbers of papers regarding CD and smoking. Numbers of IgG SE -positive patients were too small.

Response: The association between Crohn's disease and smoking is very consistent in different studies and is widely accepted; our study was not intended to address this association.

In relation to the positive association between SE IgG and smoking we agree with the reviewer and we have mentioned it in the text: a limitation of our study is the relatively small sample size, so future larger studies will be needed to confirm this observation (page 14, lines 14 to 16). However, the absence of this association in an independent sample without CD supports the notion of a specific feature of CD patients (page 14, lines 16 to 17).

- Title should be rephrased.

Response: "The Role of Salmonella enterica exposure in Chilean Crohn's disease Patients"

(2) Peer reviewer # 2

This paper represents a considerable amount of good work and merits publication, but only after the writing has been improved considerably (I have given the editor a pdf of the paper, with grammatical and spelling errors, as well as sentences with unclear meaning specified).

Response: As requested by the reviewer, we have amended spelling and grammar mistakes.

I also have a couple of concerns with your methodology: 1) it seems to me that using *S. typhimurium*

LPS in an ELISA assay to detect serum IgG molecules directed against *Salmonella enterica* could give a lot of false-negative results, given that *S. typhimurium* represents only one of ~2500 strains of *Salmonella enterica* (at the very least, you should include in your Methods section the Sigma catalog number for the LPS that was used in your study and perhaps a justification for why only one LPS was used;

Response: As suggested by the reviewer, we have included the catalog number of the LPS used in this study (page 7, line 24). We have explained in the manuscript that we used *S. Typhimurium* LPS because in Chile almost 80% of the *Salmonella*

gastroenteritis are caused by *S. Typhimurium* and *S. Enteritidis* (page 8, lines 19 to 21). A previous study has determined that the use of *S. Typhimurium* LPS in such an ELISA assay allows identification of IgG against both *Salmonella* serovars (Mette et al, *Clinical and Vaccine Immunology*, Vol. 14, No. 6, p. 741–747, 2007) (page 7, lines 20 to 21). We have modified the manuscript to include these explanations (page 7, line 24; page 7, lines 20 to 21 and page 8, lines 19 to 21).

- and the manner of defining smokers and nonsmokers seems problematic.....you define a smoker as someone who smokes at least 8 cigarettes a week, whereas a non-smoker, according to your definition, either does not smoke at all, or else, smokes less than 7 cigarettes a week (by this standard, it looks like person A who smokes 6 cigarettes a week would be placed into a separate category from person B, who smokes 8 cigarettes a week. Your data regarding the effects of smoking should be reevaluated using a more rigorous distinction between smokers and nonsmokers.

Response: The fact that raises the reviewer is applicable to any continuous variable that becomes categorical. Setting a limit in this framework can be always criticized. There is no universal criteria to define smoker versus non-smoker in Crohn's disease, however this approach is widely accepted since the classic study of Cottone M, et al (*Gastroenterology*. 1994;106:643– 648) to many other recent studies (*Ann Surg* 2005;242: 693–700; *Gut*. 2006 ; 55(2): 228–233; *BMC Gastroenterology* 2006, 6:21; *Aliment Pharmacol Ther*. 2007;25(4):429–40).

(3) Peer reviewer # 3

This is a well done and controlled study, but lacks in novelty as this is a question that multiple other studies have already aksed reaching similar conclusions. Maybe it is a confirmation of those studies in another welldefined population.


Response: Given the relevant comment made by the reviewer, we emphasize that the value of this study is 1) to help clarify the real role of Salmonella in CD in this controversial subject (page 14, lines 18 to 23) and 2) to confirm previous results in a new and well-defined population (Chilean patients) (page 12, lines 2 to 6 and page 14, lines 18 to 19) . There are no previous studies on this topic in our country or in South America.

Another comment relates to the title, the initial description of the infection and the ELISA used to establish prior exposure. While the title suggests the study is related to exposure to the Salmonella enterica serovar, the ELISA seems to be much broader in specificity as it indicates it detects Salmonella LPS.

Response: As suggested by this and other reviewer, we have modified the manuscript to indicate that we used a *Salmonella enterica* serovar Typhimurium LPS (page 7, line 24) to detect anti *Salmonella* IgG by ELISA. Patients that have been exposed to either S. Typhimurium or S. Enteritidis can be detected by this assay. We have modified the manuscript to include the reference (Mette et al, *Clinical and Vaccine Immunology*, Vol. 14, No. 6, p. 741–747, 2007) (Page 7, lines 20–21). Both serovars account for almost the 80% of the *Salmonella* gastroenteritis in Chile. We have modified the manuscript to include this explanation (page 8, lines 19 to 21).

Thank you again for publishing our manuscript in the *World Journal of Gastroenterology*.

Sincerely yours,


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