Author's response to reviews

Title: Study of the protective effect on intestinal mucosa of the hydrosoluble fiber Plantago ovata husk

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Author's response to reviews: see over
REVIEWER 1

1. Line 73, Treatments. Please indicate how the fiber was dissolved
   It has been changed in the text, the fiber was dispersed in water not dissolved

2. Why that dose of fiber was chosen? Could the authors make a comment about how the amount of fiber administered to the animals correlate with the amount human beings should uptake to get similar protective results?
   The dose of fiber was chosen taking into account the dose recommended for humans. It is the same dose as a human would take in a single administration (it has been included in the text). We chose that those in order the results obtained could be extrapolated to humans.

REVIEWER 2

Major compulsory revisions:

To identify possible mechanism of protection speculated by the authors in the discussion section, other experiments should be considered.
1) An in vitro study to examine whether fiber binds to acetylsalicylic acid (inorganic acid) and prevents intestinal injury. This is a possible mechanism as the fiber and acetylsalicylic acid were administer at a similar time within the same cannula.
   As the reviewer says, it is a possible mechanism to explain the protective action of fiber. However, in this study, to prevent the situation described by the reviewer, we prepared fiber and acetylsalicylic acid in different syringes, and cleaned cannula with water after acetylsalicylic acid administration.

2) Add histological sections with Periodic acid–Schiff (PAS) staining to determining whether intestinal mucus layer is affected by the treatments. Examination of MUC genes could also be assed to determine whether acetylsalicylic acid and/or fiber effects mucous production through altered gene expression.
   New histological sections have been added to figure 1 to include those stained with PAS, making easier caliciform cells view. As for, MUC genes evaluation, it would be an interesting study to be developed further, as our objective with this one was only to check the extent of protection conferred by this hydrosoluble fiber against acetylsalicylic acid.

3) Measuring important pro-inflammatory and anti-inflammatory cytokines levels (ie mRNA) within the intestinal tissue should also be included.
   We agree with the suggestion of the reviewer, and we think that this could be significant enough to be the object of another independent study. As we have explained above, with this study we wanted to check if Plantago ovata protected intestinal mucosa against the injurious action of aspirin.
4) **Figure 1:** High magnification images for histologically is unconvincing for epithelial injury or mitigation with fiber treatment. The lower magnification (figure 1d) provides better evidence for injury. In as such, Figure 1 should include low and high magnification images of the intestinal mucosa and importantly both treatment and control images should be presented in parallel allowing better visualization of the tissue changes. Figure 1 has been changed to include reviewer’s suggestions related to PAS staining. Moreover, low and high magnifications have been added to provide better evidence of injury and protection by fiber, and they have been presented in parallel as possible to allow better comparison of tissues.

**Minor essential revisions:**

1) **Figure 1:** Length measurement bar(s) need to be added to Figure 1
Length bars have been added to the figure.

2) A more current list of references should be added, as the most recent references in the manuscript were from 2012.
They have been updated according to the suggestion of the reviewer

3) Histologic terminology: replace the word chorion with epithelium as this term is a more accurate representation of the tissue
It has been replaced according to the suggestion of the reviewer