#Reviewer 1

We thank this reviewer for his/her critical and insightful evaluation of our manuscript.

Comment 1. "In Results, page 12, line 6, in Figure 4E, the authors should explain how the authors determine the reduction, and how the authors calculate data for the results in Figure 4E."

In 2015, Kevin A. Janes discussed in Sci. Signal. 07 Apr 2015:Vol. 8, Issue 371, pp. rs2 the limitations to use western blotting as a quantitative measurement, given this we decided to not quantify the blots. Differences in expression of proteins between groups are clearly seen in Figure 4E that is a representative blot of 2 independent experiments.

We included this information on Figure Legends section, as follows:

E, Representative western blot images of two independent experiments showing colon lysates of control and probiotic group mice; P-IKKβ, IKK, TNF-α, IL10 and alpha-tubulin.

Comment 2. "In Discussion, page 12, line 25-26, please correct mistyping."

Mistyping was corrected.

#Reviewer 2

We thank this reviewer for his/her critical and insightful evaluation of our manuscript.

Comment 1: "In the section abstract each acronym should be explained (see RT-PCR)."

Suggestion has been accepted.

Comment 2: "Considering that the probiotic group presented more tumours of smaller size (<2 mm) (P = 0.0002), could it means that probiotics slow cancer progression instead to reduce cancer development? This issue should be discussed in the section Discussion"

We agree with reviewer comment and added this paragraph in discussion:

"In aggregate these studies indicate that intestinal microbiota modulates carcinogenesis in different steps of carcinogenesis. Interestingly, the probiotic supplementation composition used in this study have its effects more pronounced in tumor initiation and promotion, as we found decreased tumour number and smaller tumour size in probiotic group."

#Reviewer 3

We thank this reviewer for his/her critical and insightful evaluation of our manuscript.

Comment 1: "Author should explain, why they choose Lactobacillus acidophilus, Lactobacillus rhamnosus and Bifidobacterium bifidum mixture for this experiment."

The choice was based on the fact that this probiotic mixture is widely used in clinical practice. In addition, literature reports anti-inflammatory effects with isolated use of these strains.

We added this paragraph in discussion:

"Prior reports showed that the isolated treatment with Lactobacillus acidophilus, Lactobacillus rhamnosus or Bifidobacterium bifidum are associated with tumour suppressive effects in colon cancer cell lines and in experimental tumour models [27-30]. Moreover, clinical studies showed that Lactobacillus and Bifidobacterium are frequently reduced in intestinal bowel disease or colorectal cancer [31]. The enrichment or depletion of different microbial strains and the change in microbial diversity is considered essential for the promotion of inflammation, proliferation and neoplastic progression [32]. Here, we used the association of Lactobacillus acidophilus, Lactobacillus rhamnosus and Bifidobacterium bifidum to assess if it can favourably alter microbiota composition."

#Reviewer 4

We thank this reviewer for his/her critical and insightful evaluation of our manuscript.

Comment 1. "Is the title really correct? Or misleading?Colon cancer-associated colitis or colitis-associated colon cancer? Which is the primary event: 1. colitis leading to 2. colon cancer (if the carcinogen is added) or 1. colon cancer and 2. dextran sulfate associated colitis as the secondary phenomenon?"

Suggestion has been accepted. We change the term colon cancer associated colitis for colitis associated colon cancer.

Comment 2. "The authors should explain the terms alpha diversity and beta diversity (for the reader not so familiar with these analyses)."

Suggestion has been accepted. For better understanding, we added an explanation, as follows:

"In this study, the alpha diversity (i.e., the number of different taxa or microbial species that could be detected in one sample) was assessed by the Shannon index and by the Chao index in the gut microbiota of the colon, and there was no difference between the control and probiotic groups. Otherwise, a significant difference was observed in beta diversity (i. e., the diversity in microbial community between different samples, accessed by the microbial composition abundances) and in the microbial composition at the genus and phylum levels. Based on these facts, it is possible to affirm that probiotic supplementation could change the structure of the microbiota."

Comment 3. "It is correct that the authors build the bridge to human pathology with coresponding data in CRC or ulcerative colitis. Can they give a suggestion which type of probiotic bacteria are most useful for therapy of inflammatory bowel diseases or for prevention of colorectal cancer (difficult question, I know)."

Despite the biological plausibility and progress in probiotic mediated CRC prevention, it is not ready for a prime time. Thus, we choose to not suggest that the specific strain composition used in our study is useful for therapy of inflammatory bowel diseases or for prevention of CRC in humans, but we suggest that clinical studies are needed. This decision was based in the number of gaps that need to be investigated, like the better dose and the time of onset and duration of treatment; and the effect of probiotic in cancer treatment.

Comment 4. "Did the authors observe any alteration of mucosal histology near the tumours? Is there a transition zone between only inflamed mucosa and malignant mucosa?"

Our data did not allow us to carry out this type of analysis convincingly. Therefore, we cannot confirm if there is a transition zone pattern.