Dear Prof. SUBRATA GHOSH,

Thank you very much for giving us the opportunity to revise our manuscript (World Journal of Gastroenterology, manuscript NO: 46080). The changes in the manuscript could be identified in track change mode.

***Reviewer #1 (code: 03260101)*:**

1. *"In addition, we also assessed a control group of 86 healthy subjects (sex, age and BMI matched), recruited from a population-based cohort study of 150 healthy subjects"* It is not clear from this sentence in Methods how exactly the 86/150 subjects were selected and whether the authors tried to minimize selection bias in the recruitment process.

1) The concerns have been addressed as follows:
We selected patients on the basis of homogeneity of baseline characteristics (age, sex and BMI) with controls using a statistical software, MatchIt (***reference***: *Ho DE, Imai K, King G, Stuart EA. MatchIt: nonparametric reprocessing for parametric causal inference. J Stat Softw 2011; 42: 1-28*). We modified the paper to better clarify this aspect (Methods section): "… and, then matched for age, sex and BMI, using MatchIt [29]. This analysis resulted in 86 patients and 86 matched controls that were enrolled for the study...".

2. "*We also identified the response (or lack of) to previous treatment about constipation symptoms*" Could the authors clarify how was this performed? Was it by screening health records or by interviewing the patients?

2) We added the following sentence in the paper (Methods section): "… also identified the response (or lack of) to past treatment about constipation symptoms by interviewing patients".

3. "*Spearman's test was performed to evaluate possible correlations of vitamin D values with quality of life scores and psychological functions*". What do the authors mean by this sentence? the significance of positive or negative correlation can be influenced by confounders. Perhaps the authors may consider to correlate Vit D serum levels with patient symptomatology.

3) We correlated the serum vitamin D levels with the scores related to health-related quality of life and psychological tests (IBS-QOL, SF12-PCS, SF12-MCS, HADS-14 A and HADS-14 D) which are expressed as continuous variables, by using Spearman's test, in patients with chronic functional constipation secondary to intestinal motility disorders. We added in the statistical analysis section: "… to health-related quality of life and psychological tests (IBS-QOL, SF12-PCS, SF12-MCS, HADS-14 A and HADS-14 D)".

4. "*For each variable included in the multivariate model, we estimated both unadjusted and adjusted odds ratios (OR), with their 95% confidence intervals (95% CI), and the level of significance (using the likelihood ratio test)*". Why did the authors calculate/ report both the unadjusted and adjusted OR?

4) For completeness we reported the results of all the analysis that we have performed.

5. "*Showed significantly reduced levels of vitamin D and higher PTH levels (for both P<0.001), even if within the normal range of values, and showed worsening quality of life (P<0.001)*".

Underlined sentence could be omitted as it does not add to the scientific observation described in the text.

5) We have deleted from the paper, as suggested, the sentence "*even if within the normal range of values".*

6.The authors do not provide the exact time point when the VIT D measurements were taken from the study's participants

6) We measured serum vitamin D levels in our patients during the period time of their enrollment, from May-June to November 2017, and the majority of them underwent to blood sampling on June-August 2017, when the sunlight exposition is high.

7. Tables with univariate/multivariate analysis should be included within the manuscript rather than been provided as supplementary materials.

7) We have excluded, as suggested, the univariate/multivariate analysis from supplementary materials and we have included it within the manuscript (Table 2).

8. I am not sure how reliably the univariate/ multivariate model can be interpreted as the authors do not provide any tabulated data on the findings of univariate analysis and subsequently chose the p value of <0.25 arbitrarily in order to construct the multivariate model.

8) Thank you for pointing out this. As suggested, we have provided tabulated data on the findings of univariate analysis and we have finally modified the Table 2 (the old supplementary table III), by excluding from the multivariate analysis the parameter of "age", which showed a *P* value >0.05 in the univariate analysis, without compromising the strong significance of vitamin D ($P<0.001$), as an independent predictor of intestinal motility disorder occurrence.

Reviewer #2 (code: 00069142):

1. It is an interesting and unique study; vitamin D deficiency is more and more involved in GI pathology and the association with constipation needs to be validated by further studies. Good english and clear discussion and conclusions.

1) Thank you for the appreciation of our study.

Reviewer #3 (code: 03478583):

1.The authors are able to show a strong correlation between Vitamin D deficiency and chronic constipation and clinical outcomes such as depression and anxiety scores. The manuscript is well-written and easy to interpret and exclusion criteria for patients seemed appropriate. The study is limited in my mind by the fact that they have not looked at other vitamin or mineral deficiencies such as Vitamin B12 or Magnesium that might also account for some of their findings. Is it possible to have additional vitamins and/or minerals measured in serum samples? That would seem to help add value to the finding that Vitamin D levels are lower.

1) Thank you for this interesting observation. We point out that we checked also for other vitamins (i.e. folate, B12, iron and ferritin) and minerals (Na+, K+, Mg2+, Ca2+) in our patients, and in the majority of them, they were within the normal range of values.

2.There is mixed literature on Vitamin D deficiency and constipation, this should be mentioned.

2) There isn't evidence both in animal and in human studies that would suggest a possible link between functional chronic constipation with slow gastrointestinal transit time and vitamin D deficiency. In the literature there is only evidence about a correlation between irritable bowel syndrome (IBS) and vitamin D deficiency, although these patients were prevalently affected by symptoms of "diarrhea" or by "alternating subtype", and only the minority of patients was categorized in the constipation variant-IBS group *(**references**: **1)** Tazzyman S, Richards N, Trueman AR, Evans AL, Grant VA, Garaiova I, Plummer SF, Williams EA, Corfe BM. Vitamin D associates with improved quality of life in participants with irritable bowel syndrome: outcomes from a pilot trial. BMJ Open Gastroenterol 2015; 2: e000052; **2)** Abbasnezhad A, Amani R, Hajiani E, Alavinejad P, Cheraghian B, Ghadiri A. Effect of vitamin D on gastrointestinal symptoms and health-related quality of life in irritable bowel syndrome patients: a randomized double-blind clinical trial. Neurogastroenterol Motil 2016; 28: 1533-44)*

3.The key question in my mind is whether slowed GI transit actually is the cause of Vitamin D deficiency, not the reverse. Is there any evidence in animals or humans that would suggest that slowed GI transit could be related to decrease Vitamin D absorption from the GI tract?

3) To the best of our knowledge, actually there isn't any study that suggests that slowed gastrointestinal transit could be related to decreased vitamin D absorption from the gastrointestinal tract; there are only studies in animals correlating vitamin D and its receptors to intestinal epithelial proliferation, but the same studies are not focused on intestinal motility.

4. Is it possible to have additional vitamins and/or minerals measured in serum samples? That would seem to help add value to the finding that Vitamin D levels are lower.

4) In this paper our primary outcome was to evaluate if serum vitamin D deficiency could be linked to chronic functional constipation secondary to delayed intestinal transit time, and consequently we investigated this relationship (and the related psychological aspects), but checking for additional vitamins and/or minerals wasn't the main objective of the study.