Dear Editor,

The authors would like to thank the Reviewers for the insightful comments, which again helped to improve the manuscript, and for the Reviewer's effort, which they greatly appreciate. The points raised by the Reviewer are addressed below.

Sincerely,

Authors

Point to point response to Reviewer's comments

Reviewer 1.

This paper investigated the correlation with polymorphisms in APOE genotype expression and clinical severity of inflammatory bowel diseases. The main findings are the lowest CRP value in UC patients with APOE e4 allele, and the youngest at first surgery but lowest PCDAI score in CD patients with APOE e2 allele. Although the clinical relevance may be modest, this paper firstly elucidated the genetic distribution of APOE polymorphism among the patients with inflammatory bowel diseases in young patients (aged 3-18). General comments Results were described in categorical variable-based orders starting with genotyping, followed by inflammation, disease localisation and behaviour, nutritional status, disease activity scales, treatment, disease severity estimates, and extraintestinal manifestations and concomitant diseases. However, going through multiple tables with different subset of patients in each section may be demanding and complicated for readers: for example, in Inflammation, serum CRP was associated with… APOEe3/e3 homozygotes (Table 3, 4, 6). Moreover, Table 3-5 compared the variables among APOE e2/e3, e3/e3 and e3/e4 whereas Table 6 compared the variables among APOE e3/e3, e2-positive, and e4-positive. The solution for the complexity may be just simply, for instance, "Table 3 shows…, Table 4 shows, or Table 6 summarised the data…. etc, so that readers can interpret the data in a step-wised manner.

Response: Thank you for this suggestion. We have altered the results section accordingly.

Specific comments

1. RESULTS, Genotyping: The authors compared the APOE genotypes with previous reports using Ref. 40-42, however inclusion of data from Ref. 52 was missing that showed similar distribution of APOE genotypes (e3/e3 = 63.0%)]

Response: Thank you for this perceptive comment. We have modified the Supplementary Data Content 2. in order to clarify that the number 86 refers to patients with *APOEε4* allele in total. Unfortunately, we could not obtain exact numbers of patients with *APOEε2/ε4, APOEε4/ε4* and *APOEε3/ε4* from authors of ref 52. Indeed in the study of Bojar et al. (ref.52) the distribution of *APOEε3/ε3* is similar to ours (62.9% vs. 62.3%; p=0.8555; OR 0.97; 95% CI 0.73-1.30). This information has been added to the manuscript. The reason for this is hard to discuss and might be linked to the fact that the previous study recruited only postmenopausal women and the method of genotyping were different to ours.

2. Table1, 3, 4, 5 and 6: The authors are advised to indicate the number of patients included the analysis at the top of the column, so that readers can estimate the number of patients in each analysis.

Response: Thank you for this comment, which will further help the readers. Number of patients has been added to all Tables mentioned.

Reviewer 2.

It is a very interesting manuscript on the hot topic of IBD epidemiology The authors try to overcome the problem of approaching disease severity using various parameters of disease activity on their pick value I think that it would be interesting if they could use one of the approved scales of disease severity Otherwise I think it would be simpler if they refer to the pick values on disease worse attack and avoid complex tables including disease presentation

Response: The authors had discussed the best approach to define severity in IBD before commencing the study. It is still a challenging task with the various aspects that have to be taken into consideration. Given that only working definitions and no formal validated or consensus definitions of mild, moderate, severe IBD currently exist regarding the course, disease burden, or related disability, we decided to present the broadest clinical picture to prevent omitting any aspects. We took into consideration that patients may have severe disease requiring aggressive therapies even if their point-in-time disease activity is not severe. This includes examples of patients with extensive steroid-dependent UC or CD refractory to immunosuppressive treatment with mild symptoms on high doses of corticosteroids. Conversely, patients may have severe symptoms without evidence of active inflammation. Although organizations such as ECCO and the American College of Gastroenterology proposed working definitions of CD and UC disease severity for clinical practice, they predominantly focus on symptoms. While discussing disease severity in either CD or UC, it is essential to think beyond clinical symptoms to include other factors critical to the patient, such as estimated days spent in the hospital, which we attempted to have.

Need for surgery is definitely a measure of disease severity but follow up is too short to refer to disease severity.

Response: We agree the follow-up is too short to refer to disease severity. We have discussed the limitation of this retrospective character of the present study in the last paragraph of discussion.

Minor genotypes should be omitted.

Response: We have changed the manuscript accordingly.

I am not sure that grouping of CD and UC is useful and apart from an initial comparison, separate presentation should be obligatory

Response: Thank you for this suggestion. UC and CD were considered separately. We have moved Table 3, comprising data for both disease entities together (UC and CD) to Supplementary Data Content 3.

Apart from hospitalisation a comparison of number of admissions is useful.

Response: The number of hospitalizations reflects the need for additional diagnosis and intensified therapy. The number of out-patient visits (if we correctly understood the Reviewer's idea), however, better reflects the parental attitude to the situation – not necessarily the clinical status of the child. Therefore, we decided to refer to the number of hospitalizations.

Albumin levels are not the only meter of nutritional status.

Response: Thank you for this remark. We have broaden the discussion section of the manuscript. We have modified the Tables 1, 3, 4 and Supplementary Data Content 3 and marked the changes in yellow. We agree albumin levels are not only the meter of nutritional status. Indeed, hypoalbuminemia at

diagnosis of UC has been reported to be associated with higher likelihood of having more than 2 courses of corticosteroids, thiopurine or anti-TNF treatment. There might also be a trend of higher risk of colectomy in patients with hypoalbuminemia [1]. In CD albumin levels were reported as marker of tight control management to avoid major adverse outcomes [2], of postoperative complications [3] and active clinical disease among CD patients [4].

[1] Khan N, Patel D, Shah Y, Trivedi C, Yang YX. Albumin as a prognostic marker for ulcerative colitis. *World Journal of Gastroenterologyl*. 2017;23(45):8008-8016.

[2] Shiga, H., Abe, I., Onodera, M. *et al.* Serum C-reactive protein and albumin are useful biomarkers for tight control management of Crohn's disease in Japan. *Scientific Reports* 2020;10, 511

[3] Müller C et al., Delta albumin is a better prognostic marker for complications following laparoscopic intestinal resection for Crohn's disease than albumin alone – A retrospective cohort study *PLOS One* 2018; 13(11): e0206911.

[4] Harris, P. et al. Association Between Serum Albumin Levels and the Rate of Active Crohn's Disease in Patients Seen at a Tertiary Care IBD Center, *The American Journal of Gastroenterology.* 2019;114, p S452

Discussion should be rewritten It should be clear if the suggestion is that apoE affects inflammation and microbiom or lipid metabolism and nutritional status From the results I can infer that the first suggestion is right

Response: Thank you for this suggestion. We have rewritten the discussion in order to focus on inflammation.