**Manuscript NO.: 79844**
**World Journal of Gastroenterology**
**Response to Reviewers**

November 12, 2022

Lian-Sheng Ma, MD
Editor-in-Chief

Dear Prof. Lian-Sheng Ma:

Thank you for the opportunity to submit a revised version of our **manuscript, "Clinical features and long-term outcomes of patients with colonic oligopolyposis of unknown etiology."** We are grateful for the valuable feedback and insights. We have responded to all the concerns below and have incorporated the suggestions made by the reviewers. Any changes made to the paper have been underlined within the manuscript. Please see below our responses to the reviewers, typed in italics. Any page numbers mentioned refer to the revised clean manuscript with tracked changes.

Reviewer #1:

Conclusion: Minor revision

1. Four patients diagnosed with invasive CRC were included in the study, while three of them were diagnosed with CRC prior to the occurrence of CPUE. Is this inclusion the correct approach? I suggest that the data of these 3 patients who had already developed CRC should be excluded. Otherwise, some genetic backgrounds or genetic backgrounds could be confounded leading to inaccurate results. This is because many genes are mutated in CRC patients.

*Author response: Thank you for this important comment. Three patients had a CRC diagnosis before developing at least ten cumulative adenomas. It is conceivable that these individuals may represent a distinct subset of CPUE patients. However, all three patients had genetic testing with large gene panels (range of 20-47 genes sequenced) that included all the polyposis-related genes, and no distinct mutations were identified. In addition, although these patients did not have 10 cumulative adenomas at the time of CRC diagnosis, all did exhibit colonic adenomas either prior to or at the time of CRC diagnosis, demonstrating that a predisposition to polyp formation was present at the same time. These findings reveal the heterogeneity of disease presentation associated with CPUE and we believe it is important to include them in the analysis.*

*We have addressed this point in the Discussion paragraph as follows:*

*"Interestingly, three patients had a CRC diagnosis before developing at least ten cumulative adenomas. However, all did exhibit colonic adenomas either prior to or at the time of CRC diagnosis, demonstrating that a predisposition to polyp formation was present at the same time. These findings reveal the heterogeneity of disease presentation associated with CPUE". (p.17, paragraph 2, lines 11-14)*

2. The authors should add some relevant references, especially for CPUE patients of different ethnicities. Because 88.5% of CPUE patients in Table 1 were non-Hispanic whites, they may not be representative of the broader CPUE patient population.

*Author response: Thank you for bringing this important point up. Our cohort may not reflect the broader CPUE population. Unfortunately, the existing literature on CPUE is very limited, and there are no reports that address CPUE in the non-white population. Information regarding the genetic epidemiology, phenotypic characteristics, and cancer risks and preventive strategies related to polyposis syndromes has been reported in mostly Caucasian individuals. We acknowledge that our observations may reflect a selection bias and that this identifies a research area of unmet need. This phenomenon may potentially reflect the lower rates of referrals for genetic counseling and testing seen in non-White populations. We have addressed this limitation in the Discussion section (p.18, paragraph 4, lines 22-23).*

*We have addressed this point in the Discussion-limitation paragraph as follows:*

*" This may result in a selection bias that could be attributable to lower rates of referrals for genetic counseling and testing in non-White populations.* [19][20]*" (p.18 paragraph 4, line 23 and p.19, paragraph 1, lines 1-2 )*

3. In Table 2, 26 patients (37%) had only the APC and MUTYH genes sequenced, but some patients with adenomatous polyposis syndrome have been reported to carry germline mutations in AXIN2, GREM1, NTHL1, POLE, POLD1, or MSH3 (Refs. 5-7). Therefore, it is difficult to accurately assess whether these 26 patients belong to the CPUE group.

*Author response: This is an important point. We acknowledge that genetic testing was not comprehensive in all patients. However, the vast majority of genetically defined cases of adenomatous polyposis are attributable to the APC and MUTYH genes. Other rare genes might potentially be involved in these patients with incomplete testing, but the frequency with which these mutations are identified is very low. For example, a study of over 3000 patients with adenomatous polyposis identified a mutation in one of these above genes in only 4 individuals (0.1%) (REF 13). We address this as one of the limitations due to the retrospective nature of the study in the Discussion section (p.18, paragraph 3, lines 16-17).*

*We have addressed this point in the Discussion-limitation paragraph as follows:*

*"However, the frequency with which mutations in these other novel intermediate-risk genes are identified is very low [13] [18] and it is unlikely that a significant number of these cases would be explained by one of these mutations". (p.18 paragraph 3, lines 17-20)*

Reviewer #2:

1. The study was designed with only 70 cases, which is too small and not enough statistical analysis can be done to draw very limited conclusions.

*Author response: Thank you for this comment. Our cohort includes 70 CPUE patients. We agree that a larger sample size would be preferable and acknowledge the limitations this places on analysis. However, the existing literature on the CPUE population is small, and our study is one of the largest to be reported. In addition, to our knowledge, our cohort is the largest in terms of the proportion of patients who had multigene panel testing. Furthermore,*

*a detailed review of demographics, clinical outcomes, and endoscopy-pathology data from a sizeable number of procedures during a more extended surveillance period enabled us to address the natural history of the condition and to reveal essential observations such as malignancy risk, long-term colonic polyposis adenoma burden, and the risk of extra-colonic involvement, which have not been previously reported. These findings expand our knowledge base and influence the clinical management of this condition.*

2. The number of references is 16, which is too small.

*Author response: Unfortunately, the literature about CPUE sub-population is quite limited. Thus, only a small number of references were cited. By addressing the other questions raised by the reviewers, we have included more references, as detailed above, and are summarized below:*

*New reference 14:*

*Long, J.M., Powers, J.M., Stanich, P.P. et al. Clinical Management of Oligopolyposis of Unknown Etiology. Curr Treat Options Gastro **19**, 183–197 (2021). https://doi.org/10.1007/s11938-021-00335-0*

*New reference 18:*

*Jelsig AM, Byrjalsen A, Busk Madsen M, Kuhlmann TP, van Overeem Hansen T, Wadt KAW, Karstensen JG. Novel Genetic Causes of Gastrointestinal Polyposis Syndromes. Appl Clin Genet. 2021 Nov 27;14:455-466. Doi: 10.2147/TACG.S295157. PMID: 34866929; PMCID: PMC863717*

*New reference 19:*

*Inra JA, Steyerberg EW, Grover S, McFarland A, Syngal S, Kastrinos F. Racial variation in frequency and phenotypes of APC and MUTYH mutations in 6,169 individuals undergoing genetic testing. Genet Med. 2015 Oct;17(10):815-21. Doi: 10.1038/gim.2014.199. Epub 2015 Jan 15. PMID: 25590978; PMCID: PMC4904772.*

*New reference 20:*

*Canedo JR, Miller ST, Myers HF, Sanderson M. Racial and ethnic differences in knowledge and attitudes about genetic testing in the US: Systematic review. J Genet Couns. 2019 Jun;28(3):587-601. Doi: 10.1002/jgc4.1078. Epub 2019 Jan 21. PMID: 30663831; PMCID: PMC8081647.*

3. The use of square brackets is inconsistent.

*Author response: We have reviewed the references and added square brackets in the for a consistent and uniform style.*

We are grateful for the insightful comments from the reviewers, and we have addressed all of the concerns raised. Please do not hesitate to reach out if there are any questions.

Sincerely,

Daniel C. Chung