October 24, 2022

Dear Lian-Sheng Ma, Founder and CEO

I am sending herewith our revised review paper entitled "**Development of EBV-associated gastric cancer: Infection, inflammation, and oncogenesis**" by Hisashi Iizasa, Andi Visi Kartika, Syntaehu Fekadu Kebede, Shunpei Okada, Afifah Fatimah, Mst. Mahmuda Khatun, Thin Myat Moe, Jun Nishikawa, and Hironori Yoshiyama, which I would like to submit for publication in ***World Journal of Gastroenterology***.

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The number ID of my invited paper is initially 03009243. However, we have received new World Journal of Gastroenterology Manuscript number,Revision-Manuscript NO: 79864.

We are looking forward to hearing from you in a short period.

Sincerely yours.

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Reviewer 1 comments

Specific Comments to Authors: *Helicobacter pylori* infection is an important risk factor in the development of gastric cancer. EBV infection is associated with many malignant tumors. The role of EBV in the development of gastric cancer is still controversial. The authors have reorganized the role of *H. pylori* and EBV infections in gastric cancer formation by reviewing both clinical epidemiological and experimental data. The authors

posed the question of whether *H. pylori* and EBV infections promote gastric cancer formation in a dependent or independent manner. The content of this article fails to clarify this problem well. Readers may be interested in the following questions, and it is suggested to add relevant content: What are the distinct molecular and clinical features of EBV associated gastric cancer? Such as the age, location, pathological characteristics and prognosis of the tumor? What are the clinical or molecular characteristics of EBV associated gastric cancer with or without *Helicobacter pylori* infection? Are there any differences between them? Is there any basic research related to the co infection of gastric epithelial cell lines with EBV and *Helicobacter pylori*? It includes simultaneous infection or successive infection.

Thank you for your very valuable comments and we have worked to resolve the issue.

1. What are the distinct molecular and clinical features of EBV associated gastric cancer? Such as the age, location, pathological characteristics and prognosis of the tumor?

   We have devided chapter 2. into A) 'Molecular features' and B) 'Clinical features' to more distinctively explain molecular and clinical features of EBVaGC.

   In 'Molecular features', on page 3, lines 18-26, we described the characteristic feature of EBVaGC, which exhibits extensive methylation in many gene promoter regions, with the exception of the MLH1 gene. The mechanism of activation of specific genes in EBV-infected cells was also presented.

   In 'Clinical features', relevance of endoscopic observation and pathological characteristics are explained on page 3, lines 29-34. The predominance of EBVaGC in men and young gastric cancer patients is discussed from line 37 on page 3 to line 4 on page 4. Prognosis of EBVaGC is explained on page4, lines 5-9.

2. What are the clinical or molecular characteristics of EBV associated gastric cancer with or without *Helicobacter pylori* infection? Are there any differences between them?

   Thank you very much for asking. It is very difficult to collect EBVaGC cases without *H. pylori* infection, because more than 95% of gastric cancer patients in Japan are infected with *H. pylori*, most EBVaGC patients are also infected with *H. pylori* [Ref. 33]. The 169 cases of noncardia gastric cancer in Korea, Japan, Poland, Mexico, and Honduras were reported to be 90% *H. pylori* positive, with a slightly higher prevalence of *H. pylori* catalase antibody positivity in EBVaGC patients [Ref.

34].

Although it is difficult to collect clinical cases, we could add one clinical study that the addition of EBV infection has been reported to increase atrophic gastritis that is caused by *H. pylori* infection [Ref. 35].

It was a bit difficult to answer the reviewer's question, but we have discussed the involvement of *H. pylori* infection in the formation of EBVaGC on page 6, lines 6-16 and 32-37, with additional references. Lines 17-31 on page 6 have been reordered slightly but remain unchanged from the first submitted paper.

3. Is there any basic research related to the co-infection of gastric epithelial cell lines with EBV and *Helicobacter pylori*? It includes simultaneous infection or successive infection.

In addition to our experimental results, several groups have conducted experiments on the relationship between EBV and H. pylori infection in the development of gastric cancer. ‘Section 5, B) Experimental observation‘ was added by increasing Ref 39 and 40, which were added and listed on page 7, lines 1-15.

Reviewer 2 comments

Specific Comments to Authors: I am pleased to evaluate your article. Good luck with your efforts. In the review, the author described the mechanism of EBVaGC from three aspects: infection of epithelial cells, induced inflammation, and carcinogenesis, which provided value for us to understand the mechanism of EBVaGC. It is a good job, but it requires some modifications to be publishable. I wish you good work. Minor observations. 1、We suggest that the authors further to discussion on the relationship between EBV and *H. pylori* on gastric cancer formation.

Thank you very much for the highly valuable suggestion. In addition to our results, several groups have also been studying the relationship between EBV and *H. pylori* infections in gastric carcinogenesis, so we cite some literature to further deepen the discussion (we have mentioned, on page 7, lines 1-15).