

ROUND 1

Dennis A. Bloomfield

Editor-in-Chief

World Journal of Clinical Cases

Dear Editors:

Thank you for inviting us to submit a revised draft of our manuscript entitled, "Fulminant amebic colitis in a patient with concomitant cytomegalovirus infection after systemic steroid therapy: A case report" to the *World Journal of Clinical Cases*.

We also appreciate the time and effort you and each of the reviewers have dedicated to providing insightful feedback on ways to strengthen our paper. Thus, it is with great pleasure that we resubmit our article for further consideration. We have incorporated changes that reflect the detailed suggestions you have graciously provided. We also hope that our edits and the responses we provide below satisfactorily address all of the issues and concerns you and the reviewers have noted.

To facilitate your review of our revisions, attached is a point-by-point response to the questions and comments delivered in your letter dated 24-Jan-2021.

Thank you for your consideration. I look forward to hearing from you.

Sincerely,

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To the Referees:

Thank you very much for your valuable comments. We are thankful for the time and effort you expended. Our responses to the referees' comments are as follows.

The revisions are highlighted in yellow.

Reviewer 1

Thank you for your comments.

1) Please describe travel history to endemic area.

Response

Thank you for this suggestion.

As you mentioned, describing the patient's travel history to endemic areas is important. Accordingly, we have added the following text to the revised manuscript: "A 68-year-old Japanese man (height: 160 cm, weight: 75 kg) was admitted to our hospital with chest pain and acute dyspnea. He worked as a truck driver and had never traveled abroad. "

2) Title should be corrected to 'Fulminant amebic colitis in a patient with concomitant cytomegalovirus infection after systemic steroid therapy: A case report

Response

Thank you for this suggestion.

We have corrected the title.

3) Page 9, Approximately 40% of patients with amebic colitis are initially misdiagnosed with inflammatory bowel disease and treated with steroids.

Response

Thank you for this suggestion.

We have corrected the sentence in question.

Reviewer 2

Thank you for your comments.

1. The specific content of the legend should be completely marked in the figure, so that readers can fully understand and learn.

Response

Thank you for this suggestion.

To make it easier for the readers to understand the figures, we have added arrows. The figure legends have also been revised as follows:

Figure 1 Chest X-ray of a 68-year-old man with chest pain and acute dyspnea on

admission. His chest X-ray shows consolidation in the right upper lobe (arrow) and bilateral congestion.

Figure 2 Colonoscopic and histopathological findings in a 68-year-old man who developed bloody diarrhea under glucocorticoid therapy. A: The colonoscopy shows multiple large ulcers in the sigmoid colon (arrow) and active colitis throughout the colon. B: The colonoscopy also shows multiple large ulcers in the rectum. (arrow) C: Neutrophilic and lymphocytic infiltration of the sigmoid colon's mucosal interstitium (hematoxylin and eosin stain, $\times 20$). D: Intranuclear inclusions of cytomegalovirus detected on immunohistochemical examination of the sigmoid colon's interstitium (arrow, $\times 20$).

Figure 3 Macroscopic and histopathological findings during autopsy of a 68-year-old man who died after intestinal perforation. A: Macroscopic findings reveal transmural intestinal necrosis and a "ragged appearance" in the rectum, descending colon, and sigmoid colon. B: Histopathological findings show full-thickness necrosis of the intestinal wall from the mucosa to the serosa. Furthermore, severe infiltration of the necrotic tissue with ameba is visible (hematoxylin and eosin stain, $\times 4$). C: A few intranuclear cytomegalovirus inclusions can be detected on immunohistochemical examination of the sigmoid colon (arrow, $\times 40$). D: Entamoeba histolytica trophozoites are visible in the submucosa (periodic acid-Schiff stain, arrow, $\times 20$).

2. In this manuscript, the diagnosis of amebic colitis was finally confirmed by autopsy. Early use of anti amebic therapy is recommended in severe colitis after steroid therapy. It is suggested that whether it can be combined with the latest

second generation sequencing is helpful for diagnosis and more accurate treatment.

Response

Thank you for this suggestion.

Accordingly, we have described the value of next generation sequencing in the revised text as follows:

Next generation sequencing is an unbiased and rapid diagnostic tool that can be helpful for accurate diagnosis and treatment.

Reviewer 3

Thank you for your comments.

Why was surgical treatment not proposed right after the diagnosis of intestinal perforation?

Response

Thank you for this suggestion.

We could not propose surgical treatment due to the patient's poor general condition.

Accordingly, the following has been added to the text:

Because of poor general condition, surgical treatment was not proposed.

ROUND 2

Thank you for your valuable comments. We are thankful for the time and effort you expended. The revisions are highlighted in yellow below and in the revised manuscript.

Reviewer 1 and Reviewer 3 Thank you for your comments.

Reviewer 2

According to the revision comments, this paper is well revised. However, for the role of second-generation sequencing in fulminant amebic colitis with cytomegalovirus infection, please briefly introduce. At the same time, combining with the relevant literature, it is emphasized that which is the best sample to be tested for higher positive rate.

Response: Thank you for this valuable suggestion. We have added relevant literature describing the use of next-generation sequencing in amoebic colitis. We could not find any literature that mentioned the complications of CMV enteritis. Therefore, in the revised manuscript, we have added the following text along with the citation. The reference cited has been added to the list of references in the manuscript. A study reported surgical cases of acute

appendicitis in HIV-infected patients; the *E. histolytica* polymerase chain reaction test result was positive in 15.8% (9/57) of the appendiceal specimens. Of the resected specimens, only two tested positive for amoebic colitis; however, a higher diagnostic rate can be expected using a next-generation sequencer[14].

14 Kobayashi T, Watanabe K, Yano H, Murata Y, Igari T, Nakada-Tsukui K, Yagita K, Nozaki T, Kaku M, Tsukada K, Gatanaga H, Kikuchi Y, Oka S. Underestimated amoebic appendicitis among HIV-1-infected individuals in Japan. *J Clin Microbiol* 2016; 55:313-320. doi: 10.1128/JCM.01757-16.