ANSWERING REVIEWERS

Sep 9, 2013

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 4949-edited.doc).

Title: Long-term follow-up of ulcerative colitis patients treated on the basis of their cytomegalovirus antigen status

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The manuscript has been improved according to the suggestions of reviewers:
1 Format has been updated

2 Revision has been made according to the suggestions of the reviewers. Below are the editor's and our corrections.

3 References and typesetting were corrected

Point-by-point response to the Reviewer 1 (No. 00044509):

Major remarks

(1) Regarding frequency and timing of CMV evaluation by the CMV antigen status.

According to the editor's suggestion, we added frequency and timing of CMV evaluation to the Materials and Methods section and the Result section.

Materials and Methods section:

CMV evaluation

…Patients were divided into two groups according to CMV antigen status: CMV-positive group and CMV-negative group. CMV antigenemia assay was performed when disease condition did not become better despite administration of immunosuppressive therapy. CMV antigenemia was usually measured once two weeks in patients with positive CMV antigen on the initial assay. Those of whom disease condition worsened after starting ganciclovir received the assay once a week.

If general condition permitted,…

Result section:

Patient characteristics

…CMV status of all the CMV-positive patients except those who underwent colectomy in a short term became negative, regardless of ganciclovir administration or not. The average period between the start of therapy and CMV antigenemia assay was 10.8±13.4 days.

The correlation between CMV antigen…

(2) Regarding bias of patients' selection and correlation between CAI and dose of corticosteroids.

According to the editor's suggestion, we added detailed information about study population, especially correlation between CAI and dose of corticosteroids to the Discussion section.

Discussion section (the 9th paragraph):

…may be almost optimal for verifying the treatment strategy for those patients. There is also a limitation about patients' selections; dose of corticosteroids at the beginning of the treatment was significantly lower in the CMV-negative group. The patients examined CMV status and enrolled in this study were suspected involvement with CMV for their refractoriness and endoscopic findings. In severer case which was suspected involvement with CMV reactivation, we would avoid using large dose of PSL not to reactivate CMV. As a result, we immediately chose second-line therapy such as CNIs, apheresis and not dose up of corticosteroids, especially in CMV-negative group, CAI of which was severer than positive group. In addition, CMV antigen was detected more frequently in male. However, I tried to stratify the results by gender and dose of corticosteroids, respectively, and we confirm that each result did not show a significant difference.

In conclusion,…

(3) Regarding negative conversion of CMV antigen status in short term, as for CMV-positive group.

According to the editor's suggestion, we added the following sentence to the Results section.

Results section:

Patient characteristics

…than those in the CMV-negative group (35 mg/day of prednisolone vs. 20 mg/day, p = 0.0003). CMV status of all the CMV-positive patients except those who underwent colectomy in a short term became negative, regardless of ganciclovir administration or not. The average period between the start of therapy and measurement of CMV antigenemia assay was 10.8±13.4 days.

The correlation…

(4) Regarding correlation between CMV negative conversion and use of ganciclovir.

As described above, CMV disappeared in all the 40 patients regardless of GCV administration or not. However in not only short-term but also long-term, ganciclovir could improve the prognosis of UC patients. According to the editor's suggestion, we added the following sentence to the Discussion section.

Discussion section (the 3rd paragraph):

…In contrast, however, no significant difference in colectomy rate was observed between patients with and without CMV, even in the short-term. ~~One quite reasonable explanation for this is that ganciclovir could contribute to inducing remission in the CMV-positive patients through the correct selection of CMV "disease" among relapse of UC. In fact, we showed that the administration of ganciclovir was the only predictive factor of avoidance of colectomy.~~ In this context, the CMV antigenemia assay could reliably detect CMV "disease" which requires antiviral therapy. In our finding, use of ganciclovir is not always correlated with CMV negative conversion in short-term but a predictor of avoidance of colectomy in long-term. These suggest that use of ganciclovir exerts clinical effectiveness in long-term follow-up, e.g., avoiding exacerbation and/or relapse after remission, for a portion of patients with CMV.

In term of diagnosis regarding…

Point-by-point response to the Reviewer 2 (No. 00068316):

Major remarks

(1) Regarding selection bias and stratification by sex and dose of corticosteroids.

In this regard, we responded at the comment (2) of the reviewer1.

(2) Regarding the sum of percent of short term remission rate in CMV positive group.

According to the editor's suggestion, we correct the percent to one decimal place in short term remission rate. Similarly, we correct the notation about percent to one decimal place through the manuscript.

(3) Regarding analysis of remission rate between two groups limited to CMV-positive with GCV and CMV-negative without GCV.

According to the editor's suggestion, we added the following sentence to the Results section.

Results section:

Short-term remission rates according to CMV antigen status

…in the CMV-negative group (21 days vs. 16 days, p = 0.009, Mann-Whitney U-test). In addition, we analyzed remission rate from the starting day of the remission-induction therapy between the two groups; one group was limited to CMV-positive patients administered ganciclovir and another was CMV-negative patients not administered ganciclovir. These curves showed the better clinical course in the CMV-negative group, too (p = 0.03, log-rank test).

(4) Regarding the reference about pathogenicity of CMV in UC patients with flare-up.

According to the editor's suggestion, we added the following reference to the Discussion section.

Discussion section (the 3rd paragraph) :

…short-term. This result is consistent with those of previous reports[9,27]. In contrast…

(5) Regarding some grammar errors and missing spaces.

According to the editor's suggestion, we sent the professional English language

editing company. Please find attached certification.

Point-by-point response to the Reviewer 3 (No. 01588775):

Comment 1.

Regarding correlation between refractoriness to remission-induction treatment and reduction of corticosteroids in CMV-positive patients.

According to the editor's suggestion, we performed multivariate analysis for short-term remission. However, the dose of reduction of corticosteroids was not a significance factor in the short term (OR = 0.94; 95%CI, 0.41-2.39, p = 0.90). So we added the following sentence to the Discussion section.

Discussion section (the 5th paragraph):

In addition, our results indicated that reduction of the dose of corticosteroids and immunosuppressive therapies were not significant factors in multivariate analysis for avoiding colectomy both in the short and long-term. This suggests…

Comment 2.

Regarding table 2; correlation between immunohistochemistry and CMV antigenemia assay.

According to the editoor's suggestion, we removed table 2 and added the following sentence to the Result section.

Results section:

Patient characteristics

…the CMV antigenemia assay. Of the 23 patients in the CMV-positive group, 9 (39.1%) were positive for IHC. On the other hand, 25 (96.2%) of the 26 patients in the CMV-negative group

were negative for IHC. The results of the CMV antigenemia assay were closely correlated with IHC of inflamed colon mucosa for CMV (p = 0.003, Fisher's exact test)….

Thank you again for publishing our manuscript in the *World Journal of Gastroenterology*.

Sincerely yours,

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