

EDITORS IN CHIEF:

Professor Damian Garcia-Olmo, Stephen Strom and Andrzej Tarnawski

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RE: MS No. 36404 "Early Gastric Cancer Frequently Has High Expression of KK-LC-1, a Cancer-Testis Antigen" by N. Futawatari et al.

Dear Editors:

Thank you for your letter dated October 20, 2017. The comments of the two reviewers have been very helpful to improve our manuscript. We have responded all comments, performed additional data along with the suggestions and revised the manuscript based on their suggestions. We added the I) image of KK-LC-1 amplicon of 342bp in Figure 1 and II) the data about expression of CTAs in intramucosal, submucosal 1 and submucosal 2 gastric cancer into Table 2. We would like to thank you for allowing us to resubmit the revised manuscript.

I hope that the revised manuscript can again be considered for publication in the Journal. Thank you in advance for your kind consideration.

Sincerely yours,

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For the comments of the reviewer 1 (Reviewer's code: 03656586):

We are grateful to your appreciation of our study.

1. How to get early cancer specimens

We got the early cancer specimens from the stomach after surgical resection, not endoscopic operation. We aborted to collect tumors which size were less than 10mm because collection from them might impinge on the pathological diagnosis.

2. What are the advantages and disadvantages compared KK-LC-1 with the currently recognized signs of gastric cancer tumor markers?

We think below

Advantage: KK-LC-1 was known as a target for cancer immunotherapy so that the detection of KK-LC-1 will direct to companion diagnosis for cancer immunotherapy.

Disadvantage: The assay of KK-LC-1 detection is only biopsy. The diagnosis system to detect KK-LC-1 is not established. It will need the diagnosis system to detect more easily such as the detection from serum as well as other tumor markers.

3. Just use an experimental method to verify(PCR), whether it is too thin?

We added the figure 1 including the 342bp amplicon of KK-LC-1 by PCR to verify its intensity for positive assessment.

For the comments of reviewer 2 (Reviewer's code: 00502831)

Thank you very much for your valuable comments.

Answers to specific comments

#1. How is statistical comparison between tumor size and the expression of KK-LC-1?

We statistically analyzed the tumor size between positive and negative of

each CTA, but no significance were found. We added them into Table 1.

#2. The authors reported that the KK-LC-1 expression rate was high even in early gastric cancer. So the authors should describe about the KK-LC-1 expression in intramucosal, SM1 and SM2 cancer, separately.

We added the CTAs expression including KK-LC-1 in intramucosal, SM1 and SM2 cancer in part of Table 1. Although the number of patient was small, the tendency that higher rate of KK-LC-1 expression was less of tumor depth was found. In near future, we will report the comparison of KK-LC-1 expression in the tumor depth.

#3. How about cases with pseudo-positive and pseudo-negative of the KK-LC-1 expression? The authors should describe pseudo-positive and pseudo-negative cases in detail.

We think pseudo-positive cases would be few because we assessed the visible 342 bp amplicon, in which we haven't experienced the contamination of other sequences, as positive cases. However, pseudo-negative cases might be included in this study because we couldn't distinguish the specimens included minority of tumor cells. We added them in page 10 line 17 to 24.

The critical comments and useful suggestion have helped to improve our paper. We have attempted to answer each of the questions raised.

Thank you in advance for your kind consideration of our revised version.