

Name of Journal: *World Journal of Gastroenterology*

Manuscript NO: 38688

Title: Olfactomedin-4 in digestive diseases: a mini-review

Dear Editor,

Thank you very much for your email dated 31 March 2018, and the valuable comments of the two referees and editors. Based on the comments, we have made extensive modification on the original manuscript. Here, we attached the revised manuscript in which major changes made to the text were underlined so that they may be easily identified. A list of answering every comment from the referees was also enclosed. We hope that the revised manuscript is acceptable for publication. Thank you!

Yours Sincerely,

Chengfu Xu, M.D.

Department of Gastroenterology

The First Affiliated Hospital, College of Medicine, Zhejiang University

79 Qingchun Road, Hangzhou 310003, P. R. China

Tel: +86-571-87236532; Fax: +86-571-87236611

E-mail: xiaofu@zju.edu.cn

Replies to the Review #1

Comment: This is a very well written review paper which analyze the available literature data concerning the expression, function and regulation pathway of Olfactomedin-4 (OLFM4) as a potential diagnostic marker and a therapeutic target in *H. pylori* infection, inflammatory bowel disease and gastrointestinal malignancies. The authors give the extensive overview about the expression, function, regulation peculiarities of OLFM4 protein by different digestive diseases and discussed the role of OLFM4 as potential marker of disease. The authors have reviewed and analyzed a sufficient amount of literature (53 references). This paper makes a contribution to studies concerning the better understanding of the role and function mechanisms of OLFM4 in pathogenesis of gastrointestinal disorders.

Reply: Thank you very much for your valuable work and positive comments on our manuscript!

Replies to the Review #2

Comment 1: In which cell district is OLFM4 expressed? In membrane, cytoplasm or nucleus?

Reply: We apologize for the confusion! OLFM4 is a glycoprotein that could be expressed in membrane, cytoplasm, nucleus, mitochondria and granules of mature neutrophils [1-3]. We have added this information in the revised manuscript on page 5, line 9-10.

Page 5, line 9-10: OLFM4 can be expressed in the membrane, cytoplasm, nucleus, mitochondria, and mature neutrophil granules.

Comment 2: It is unclear what is the primary function of OLFM4.

Reply: Thank you very much for this comment! The precise functions of OLFM4 in digestive diseases have not been fully understood. The primary function of OLFM4 in gastrointestinal malignancies is associated with its role

as an antiapoptotic factor that promotes the tumor growth [2]. In addition, OLFM4 downregulates innate immunity against *H. pylori* infection^[4] and affects anti-inflammatory function in IBD^[5]. We have added this information in the revised manuscript on page 5, line 16-20.

Page 5, line 17-21: The primary function of OLFM4 in gastrointestinal malignancies is associated with its role as an antiapoptotic factor that promotes the tumor growth. In addition, OLFM4 downregulates innate immunity against *H. pylori* infection and affects the anti-inflammatory function in IBD.

Comment 3: Authors should clarify whether the over-expression of OLFM4 in *H. pylori* and other diseases is due to increased production by epithelial cells or neutrophils/lymphocytes.

Reply: Thanks very much for this comment! Recent studies have revealed that OLFM4 expression is up-regulated in the neutrophils, macrophages and epithelial cells after *H. pylori* infection. Thus, the over-expression of OLFM4 in *H. pylori* infection is due to the direct action on epithelial cells as well as activated infiltrating of neutrophils and macrophages ^[4]. We have added this information in the revised manuscript on page 6, line 10-14.

Page 6, line 10-14: The expression of OLFM4 is upregulated in neutrophils, macrophages, and epithelial cells after *H. pylori* infection, which suggests that overexpression of OLFM4 upon *H. pylori* infection is due to its direct action on epithelial cells as well as to activation of neutrophil and macrophage infiltration.

Comment 4: Are there any evidences that *H. pylori* eradication leads to normalization of OLFM4 levels?

Reply: Thank you very much for this comment! There is no study examining whether *H. pylori* eradication leads to OLFM4 levels change. It is worth to

perform further studies to clarify this issue. We have added this information in the revised manuscript on page 6, line 8-10.

Page 6, line 8-10: However, further study is warranted to determine whether eradication of *H. pylori* leads to the normalization of OLFM4 levels.

Comment 5: Pancreatic cancer may occur in a background of chronic pancreatitis. Therefore, are there any studies examining OLFM4 expression in chronic pancreatitis or during acute pancreatitis flares?

Reply: Thank you very much for this comment! There is no study clarifying the correlation between OLFM4 expression and either chronic pancreatitis nor acute pancreatitis. It is an interesting area that is worth further investigations. We have added this information in the revised manuscript on page 11, line 22.

Page 11, line 25-27: Pancreatic cancer may occur in a background of chronic pancreatitis. Whether OLFM4 is associated with chronic pancreatitis or acute pancreatitis flares is worth further investigations.

Comment 6: In conclusion, the present manuscript seems to be too much descriptive. For this reason, a table summarizing the effects of OLFM4 in the various gastrointestinal diseases may make the article more appealing for the readers.

Reply: Thanks for your advice about adding a summarizing table. We have added it (Table 1) below the conclusion section in the revised manuscript on page 23.

Page 23: Table 1 is added.

Replies to the Editors

Comment 1: Please add some figures and tables in the main text

Reply: Thank you very much for your valuable suggestion! We have added a figure and a table in the revised manuscript on page 22 and page 23.

Page 22: Figure 1 is added.

Page 23: Table 1 is added.

Comment 2: Audio core tip.

Reply: We have submitted an audio core tip as requested.

Comment 3: Please check and confirm that there are no repeated references.

Reply: We have checked and confirmed that there is no repeated in reference section.

Comment 4: Please provide language certificate letter by professional English language editing companies (Classification of manuscript language quality evaluation is B).

Reply: The manuscript has been edited by native English-speaking editors at American Journal Experts. A language certificate is provided by AJE as requested.

Comment 5: You need to provide the grant application form(s) or certificate of funding agency for every grant, or we will delete the part of "Supported by...".

Reply: We have provided the certificate of funding agency as requested.

References

- 1 Liu W, Lee HW, Liu Y, Wang R, Rodgers GP. Olfactomedin 4 is a novel target gene of retinoic acids and 5-aza-2'-deoxycytidine involved in human myeloid leukemia cell growth, differentiation, and apoptosis. *Blood* 2010; **116**(23): 4938-4947 [PMID: 20724538 DOI: 10.1182/blood-2009-10-246439]
- 2 Zhang X, Huang Q, Yang Z, Li Y, Li CY. GW112, a novel antiapoptotic protein that promotes tumor growth. *Cancer Res* 2004; **64**(7): 2474-2481

[PMID: 15059901]

- 3 Liu W, Yan M, Liu Y, McLeish KR, Coleman WG, Jr., Rodgers GP. Olfactomedin 4 inhibits cathepsin C-mediated protease activities, thereby modulating neutrophil killing of *Staphylococcus aureus* and *Escherichia coli* in mice. *J Immunol* 2012; **189**(5): 2460-2467 [PMID: 22844115 DOI: 10.4049/jimmunol.1103179]
- 4 Liu W, Yan M, Liu Y, Wang R, Li C, Deng C, Singh A, Coleman WG, Jr., Rodgers GP. Olfactomedin 4 down-regulates innate immunity against *Helicobacter pylori* infection. *Proc Natl Acad Sci U S A* 2010; **107**(24): 11056-11061 [PMID: 20534456 DOI: 10.1073/pnas.1001269107]
- 5 Liu W, Li H, Hong SH, Piszczek GP, Chen W, Rodgers GP. Olfactomedin 4 deletion induces colon adenocarcinoma in *ApcMin/+* mice. *Oncogene* 2016; **35**(40): 5237-5247 [PMID: 26973250 DOI: 10.1038/onc.2016.58]