

Dr Ze-Mao Gong
Science Editor,
World Journal of Gastroenterology

Dear Dr Ze-Mao Gong,

Thank you for considering our invited manuscript "*Helicobacter pylori*: a foodborne pathogen?" (Manuscript number: 39636 Column: Review) for publication in World Journal of Gastroenterology. We also thank the reviewers for their thoughtful comments, which have been extremely helpful for the final improvement of our manuscript. The manuscript has been modified according to your indications, and below are the point-to-point answers to the corrections required by reviewers. The manuscript was revised by a language editing service and a language editing certificate has been attached. We have added a list of abbreviations as suggested by reviewer 00055108.

We hope the revised version is acceptable for publication. All the revisions are written in red.

Kind regards,

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Editor's comment

We have done everything

Reviewer's code 02535288**Specific comments to authors**

This work by Drs. Quaglia et al., have reviewed the evidence of foods and water in the transmission of H. pylori to humans and the methods for detecting pathogens in foodstuffs and water. Numerous published works have tried to reveal the routes of how H. pylori infect human being, current work could not fully reveal the routes of infection, therefore further works in these areas will be a welcome. This work is therefore interesting with merit. The manuscript appears well organized and written, but also appears lengthy. Adding a few charts or cartoons may strengthen the overall quality of work and might be a better form of presentation for reader to understand available data and hypothesis. The work may require revision with supporting data to strengthen the claim and point out directions for future investigation to guide readers. Authors are encouraged to revise and make the manuscript more concise and attractive.

We thank the reviewer for his kind comments. We have added a chart and 3 tables to the main text to make the manuscript more concise and attractive as suggested. The tables included data that were not described in the text but only commented, to reinforce the indications for future investigations, as reported in the conclusions section.

Reviewer's code 00503628**Specific comments to authors**

This is timely and well-written review on an important topic regarding H. pylori transmission. It nicely summarizes work that has been done in that area.

We thank the reviewer for his kind comments regarding our work.

Reviewer's code 01557050**Specific comments to authors**

Drs. Quaglia and Dambrosio, et al reviewed 'Helicobacter pylori: A FOODBORNE PATHOGEN? The manuscript is well-presented. The reviewer has some comments. The reviewer understood the

existence of HP in water, foods and so on. In clinically, the reviewer is interested in the prevention of HP infection from foods. Then, we should establish hygiene methods in each countries. Furthermore, the reviewer would like to know that the correlation between Cag A and carcinogenesis might be also infection from foods or not.

We thank the reviewer for his kind comments.

In European Community countries, food hygiene and safety are regulated by the Hygiene Package, which sets some rules on hygiene and official controls of food products.

The Hygiene Package aims to ensure a high level of protection of human health by ensuring safe and healthy food. The rules define specific hygiene requirements valid to all foods and risks, such as the presence of *Helicobacter pylori* in food.

Furthermore, prevention must also include consumer awareness in observing basic hygiene standards. In developing countries, governing bodies should place a consumer awareness campaign to adopt good hygiene standards during food preparation and consumption.

Concerning the correlation between food and carcinogenesis due to the presence of the *cagA* gene in strains isolated from foods and water, as far as I know, there are no data in the literature.

Reviewer's code 02441021

Specific comments to authors

EVIDENCE SUPPORTING THE ROLE FOR WATER IN THE TRANSMISSION OF H. pylori TO HUMANS Corrections: • Contaminated water can transmit diseases such as diarrhoea, cholera, dysentery, typhus and polio [82]..... Contaminated water can transmit diseases such as diarrhoea, cholera, dysentery, typhoid and polio [82]. • It has been estimated that H. pylori colonizes that more half of the world's population,..... It has been estimated that H. pylori colonizes more than half of the world's population, Occurrence of H. pylori in water Corrections: • Acanthamoeba, Naegleria, Vermamoeba or Balamuthia,.....should be in I talicsAcanthamoeba, Naegleria, Vermamoeba or Balamuthia,

We thank the reviewer for his kind comments. We have corrected the manuscript as suggested. All corrections are in red.

Reviewer's code: 00183445

Specific comments to authors

1. *This is a good and valuable article focusing on Helicobacter pylori a gastric pathogen as a potential foodborne pathogen. The article presents an overview of current knowledge about phenotypic characteristics, growth requirements of H. pylori, the reservoirs, transmission modes, including animals, water and foodstuff. Furthermore, the panel of methods used for the detection of H. pylori in food and water has been described. Much attention has been devoted to the role of coccoid forms in the transmission of infection. Specific comments.*

Concerning human-to-human transmission I propose to add: H. pylori may be a sex transmitted pathogen (1, 2) and it may lead to fibrocystic breast changes (3). The oral cavity can be primarily colonized by H. pylori and this can be linked with later gastric infection (4).

1. Singh V et al, Helicobacter pylori: evidence for spouse-to-spouse transmission. J. Gastroenterol Hepatol 1999, 14:519-522. 2. Schutze K et al, Helicobacter pylori reinfection with identical organisms: transmission by the parents' spouses. Gut 1995, 36:831-833. 3. Kast RE. Some fibrocystic breast change may be caused by sexually transmitted H. pylori during oral nipple contact: Supporting literature and case report of resolution after gut H. pylori eradication treatment. Med Hypotheses 2007, 68:1041-1046. 4. Yee JKC. Are the view of Helicobacter pylori colonized in oral cavity an illusion?. Nature. Experimental & Molecular Medicine 2017 49, e397, DOI:10.1038/emm.2017.225

We thank the reviewer for his kind suggestions. We have added the sentences suggested, as well as the references in the main text and in the references list. All corrections are in red.

2. *Chapter Microbiological Characteristics Phenotypic characteristics Page 5 "Bacteria in the VBNC state maintain their metabolic activity and pathogenicity and the ability to return to active regrowth conditions [37, 38]. This ability of H. pylori to overcome stress conditions...". The property of H. pylori to return to active regrowth conditions has not been proven. It should be clearly stated. Data in 37 and 38 articles relate to other gastric pathogens.*

We thank the reviewer for his kind comments regarding our work. The sentence has been corrected as suggested. All corrections are in red.

3. *Survival of H. pylori in foodstuff Page 8 "However, a study on survival of H. pylori in artificially contaminated spinach showed that the bacterium is able to survive for up to 6 days in VBNC forms that are still viable and virulent despite their lack of cultivability [54]" and Methods for the*

detection of H. pylori in foods and water Page 15 "....however, it has been hypothesized that VBNC forms may still be infective [131, 132] thus representing a potential microbiological risk for consumers' H. pylori VBNC are not infectious in the sense that H. pylori spiral forms are, which can multiply. However, during chronic H. pylori infection, VBNC forms can often arise in unfavorable conditions of inflammatory milieu and can be a source of virulence factors involved in the pathogenesis process: induction of gastric barrier dysfunction and the maintenance of pathologic inflammatory response. This should be more emphasized.

We thank the reviewer for his kind comments. We have corrected the sentences as suggested. All corrections are in red.

Reviewer's code: 00055108

Specific comments to authors

The major issue to this well-written manuscript is that it do not reveal any information regarding your method of search for appropriate literature to include in this review. Please consider to do so.

We thank the reviewer for his kind comments regarding our work. We have added the chapter "Literature search" to our manuscript.

Comment D1: Consider also to include "waterborne" in the title, it will suit the purpose?

We thank the reviewer for his kind suggestion. The title of the review was previously accepted by the Editor, so I do not know if I can change it as suggested. If this is possible, I will be happy to do it. We have added "water" to the running title.

Comment D2: Consider to make a liste of abbrevations

We thank the reviewer for his kind suggestion. The list of abbreviations has been added to the end of the manuscript.

Comment D3: Core tip. Two long – consider a 50% reduction

We thank the reviewer for his kind suggestion. We have removed the first sentence of the core tip.

Comment D4: Even though it's not a systematical review please consdider to add a sentecne how/and were you gained your information.

We thank the reviewer for his kind suggestion. We have added a chapter to the manuscript entitled "Literature search" after the Introduction.

Comment D5: Obs – english – consider rephrasing?

We thank the reviewer for his comment. We have corrected the sentence.

Comment D5: The typical routine diagnostic tests are the indirectly rapid urease test (different kits) or the Hp fecal antigen test – furthermore the 13/14C urea breathing test. Gastric biopsy for bacterial culture is not used as a routine diagnostic, in many cases it is explored when there is treatment failure – antibiotic resistance.

We thank the reviewer for his kind comment. To better explain the use of *H. pylori* culture in clinical diagnostics, we added a sentence.

Comment D6: Meaning? Definition?

We thank the reviewer for his kind suggestion. We wrote water activity instead of a_w , and we have added this word to the abbreviation list.

Comment D7: Expressed as a_w above, please be consistent.

We thank the reviewer for his kind suggestion. We wrote a_w instead of water activity.

Comment D8: This is important, consider to also write it even earlier in the manuscript. It's a general information regarding Hp

We thank the reviewer for his kind suggestion. We have added a sentence in the Introduction chapter.

Comment D9: "was also investigated" English?

We thank the reviewer for his kind suggestion. All the manuscript has been revised by a language editing service.

*Comment D10: "It has been estimated that *H. pylori* colonizes that more half of the world's population, and contaminated water is mentioned as one of major causes" English.*

We thank the reviewer for his kind comment. All the manuscript has been revised by a language editing service.

Comment D11: consider "demanding" or another word.

We thank the reviewer for his kind correction. We have replaced *exacting* with *demanding* as suggested.

Comment D12: Consider the need of a short explanation

We thank the reviewer for his kind comment. A short explanation is in the chapter MICROBIOLOGICAL CHARACTERISTICS- Growth requirement.

Comment D13: "As is well known, the main limit of PCR assays is their inability to distinguish live organisms from dead organisms. PCR techniques can, however, be used to screen water and

foodstuffs, thus making it necessary to use conventional isolation methods only on those samples that test positive by PCR.” Consider to reveal this information earlier in the manuscript

We thank the reviewer for his kind suggestion. This sentence is useful at this point of the manuscript to explain what is written later. Therefore, revealing this information earlier in the manuscript would take importance away from what is described later.

Reviewer’s code: 02444986

Specific comments to authors

1. this is a review of literature for transmission of h. pylori 2. authors emphasised foods as a possible route of transmission for h. pylori transmission. 3. there is no clear conclusion on whether h.pylori can be transmitted by oral-decal root. Authors did not make any comment on the future studies to clarify this question.

We thank the reviewer for his kind comments. In the literature, contamination of water and food has been attributed to poor environmental hygiene conditions. In fact, a greater incidence of pathology and greater bacterial isolation of the pathogen from food and water occur in developing countries. In our opinion, data on the source of primary contamination, i.e., the reservoir, are poor. Therefore, we have added a sentence to clarify what future studies are needed to verify the source of contamination of water and food.

Reviewer’s code: 00538725

Specific comments to authors

It's an informative review on the potential role of H. pylori as foodborne pathogen for human diseases. The contents considered the survival environment, transmission passage, and detection methods of H. pylori in food and water. The literature review work was adequate and up-to-date. It can be recommended for publication.

We thank the reviewer for his kind comments regarding our work.

